

Discrete Stochastic Model for the Generation of Axonal Trees

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Outline

- 1 Introduction
- 2 Model Description
- 3 Validation
- 4 Conclusions

Introduction: Motivation

- During the development of the nervous system, neurons extend their axons to form neural circuits
- Many of the mechanisms involved in the axonal biogenesis process are still not completely understood
- Realistic simulation frameworks could provide an insight into these processes

Introduction: Motivation

- Axons are guided to their target by their growth cones (GC)
- GC sense chemical signals present in their environment
- The paths taken by the GC depend on these signals
- Once an axon reaches its destination, it generates branches

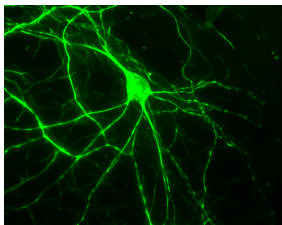


Figure: Population of neurons creating a neural circuit (from <http://www.encorbio.com>).

Introduction: Objective

- Develop a model for the generation of realistic axonal trees
- All the model parameters should be estimated from static images (confocal microscopy).
- In particular, we work with axons of *Drosophila* flies (wild and mutant)
- Use the parameters to characterize different populations (wild and pathological)

Introduction: Motivation

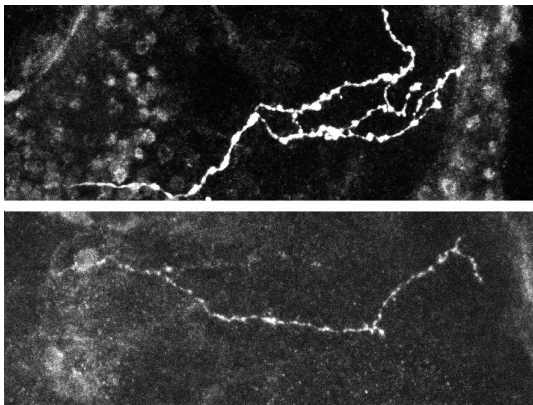


Figure: Wild (top) and Mutant (bottom) (MIP)

Model Description

- 2D discrete stochastic model for the simulation of axonal biogenesis
- Considers 2 main biological processes:
 - Growth process: models the elongation and shape
 - Bifurcation process: models the generation of branches
- Both processes are considered to be independent from each other

Model Description: Growth process

- The number of points N of a filament is determined using a Gaussian distribution $\mathcal{N}(\mu, \sigma^2)$
- The shape is determined using a third order Markov Chain that depends on the elastic properties of the neurites and on the external attraction field (parameters of the model)

Model Description: Growth process, Shape

- Given a point n_t on a path, n_{t+1} depends on:
 - Relative positions of n_{t-1} , n_{t-2}
 - External field in n_t , given by a vector
- Two main cases are defined (plus all the possible rotations)

Model Description: Growth process, Shape

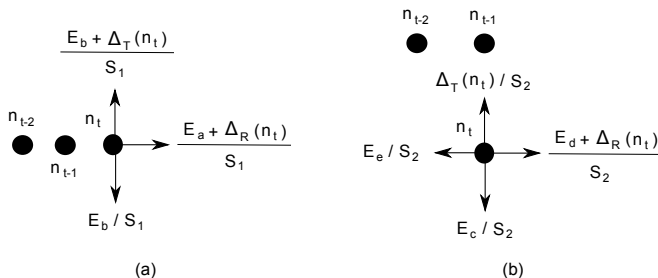


Figure: Conditional probabilities $P(n_{t+1} | n_t, n_{t-1}, n_{t-2})$

$$\begin{cases} S_1 = E_a + 2E_b + \Delta_R(n_t) + \Delta_T(n_t) \\ S_2 = E_c + E_d + E_e + \Delta_R(n_t) + \Delta_T(n_t) \end{cases} \quad (1)$$

Model Description: Bifurcation process

- Bifurcation process is defined by the probability of branching P_b on each point
- $P_b(n)$ is piecewise constant during time
- Branches grow independently of the main axon following the same model.

Model Description: Parameter estimation

- Model parameters: $(\mu, \sigma^2, P_b, E_a, E_b, E_c, E_d, E_e)$ and the external field $(\Delta_x(u, v), \Delta_y(u, v))$, for $(u, v) \in L$.
- All estimated from real images of axonal trees

Model Description: Parameter estimation, μ and σ

- Length of paths with normal distribution:

- $\hat{\mu} = \frac{\sum_{a \in A} l_a}{\text{card}(A)}$

- $\hat{\sigma}^2 = \frac{\sum_{a \in A} (l_a - \hat{\mu})^2}{\text{card}(A)}$

- where A is a set of axonal trees and l_a , $a \in A$ the length (i.e., number of pixels) of the main branch of axon a .

Model Description: Parameter estimation, P_b

- $$\hat{P}_b = \frac{1}{\text{card}(A)} \sum_{a \in A} \frac{N_b(a)}{I_a}$$

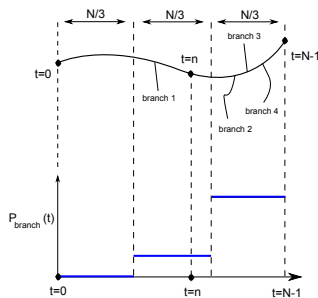


Figure: P_b estimation

Model Description: Parameter estimation

- For each configuration of the past (i.e, n_{t-1}, n_{t-2}) we obtain equations
- In total, 48 equation are considered (when we take into account all the possible rotations)
- The equations relate the remaining model parameters with the number of times the different possible configurations appear in the images

Model Description: Parameter estimation

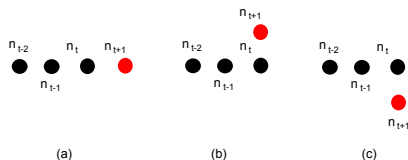


Figure: Example of possible configurations

$$\begin{cases} \frac{N_1}{N_T} = \frac{\hat{E}_a + \hat{\Delta}_R}{\hat{S}_1} \\ \frac{N_2}{N_T} = \frac{\hat{E}_b + \hat{\Delta}_T}{\hat{S}_1} \\ \frac{N_3}{N_T} = \frac{\hat{E}_b}{\hat{S}_1} \\ N_T = N_1 + N_2 + N_3 \end{cases} \quad (2)$$

where $N_{1,2,3}$ are the number of times that the configurations are present in an axon (assumption: $\hat{\Delta}_R$, $\hat{\Delta}_T$ constant)

Model Description: Parameter estimation

- The system of 48 equations is solved with Least Mean Squared
- The parameters are estimated using sliding window scheme (to allow variation in the chain parameters)

Validation

- Validated on 53 confocal microscopy images of *Drosophila* axons
- Both normal and mutated neurons were considered (18 normal, 21 type 1 mutant and 14 type 2 mutant)
- Each image has 2 channels
 - Channel 1: Axonal tree
 - Channel 2: Brain structure in which axons are developing
- Images were registered (using the second channel) and the axons segmented by a biologist

Validation

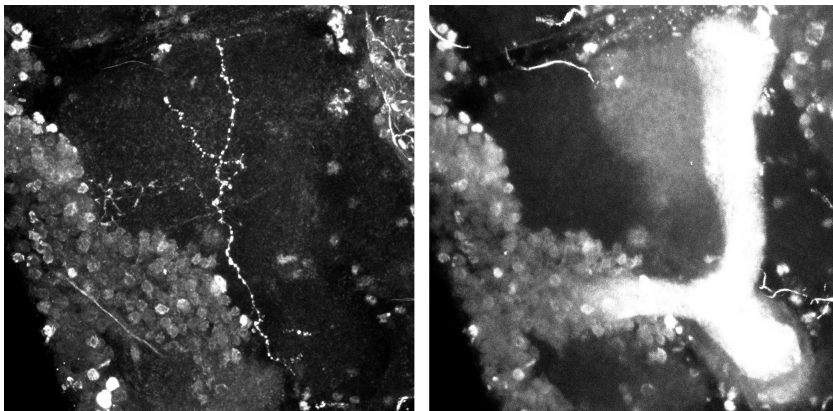


Figure: Channel 1 (left) and 2 (right)

Validation

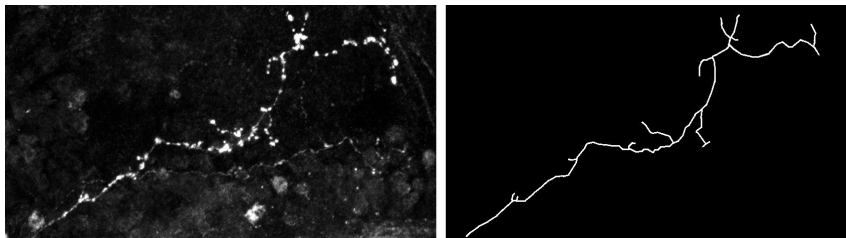


Figure: Original (left) and manual segmentation (right)

Results

- The parameters were estimated and averaged between images to obtain representative values for each population
- Synthetic axons were generated and compared with real trees for each population

Results: Normal tree

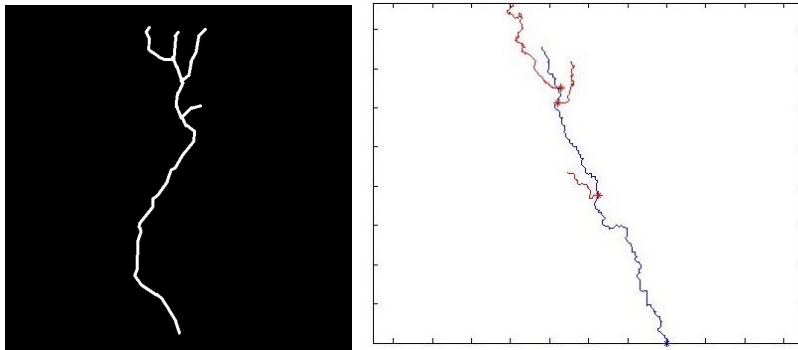


Figure: Real (left) and synthetic (right) trees

Results

- We have calculated a scalar field (E_a/E_b) and a vector field (Δ) for each image
- Field extrapolation (using Gaussian Markov Random Field) to obtain non sparse fields (2 per population)
- Fields are plotted on the (registered) neuronal structure in which axons are developing (channel 2)

Results

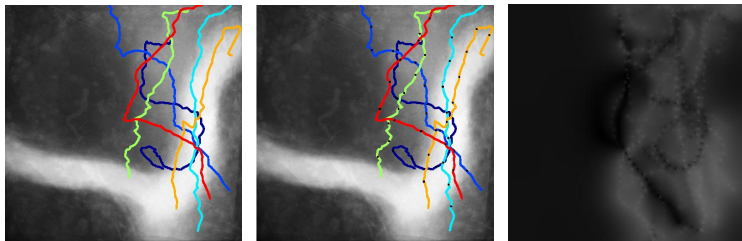


Figure: Axons on registered image (left), points with parameters estimated (middle) and extrapolated field (right).

Results: Δ field normal population

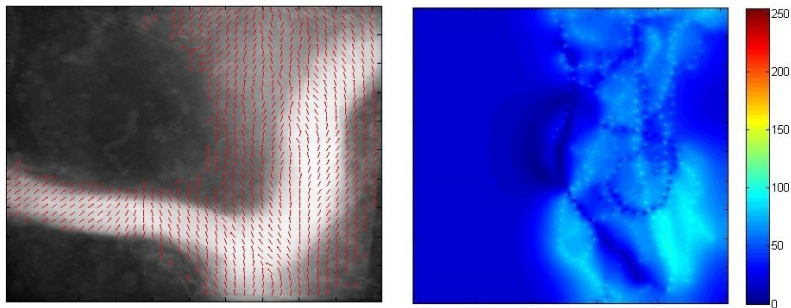


Figure: Direction (left) and norm (right) of the Δ field for the normal population.

Results: Δ field

- Qualitative comparison between the Δ fields of different populations suggest no relevant difference between them
- Field points towards the target area (top right corner of the image)
- Field norm is stronger at the starting point of the axons (bottom right section) and weaker near the target area.

Results: E_a/E_b scalar fields

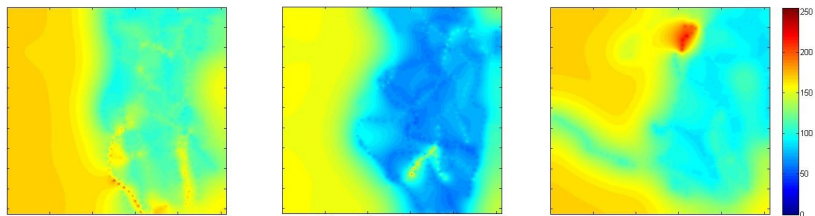


Figure: E_a/E_b scalar field for the normal (left), mutant type 1 (middle) and mutant type 2 (right) populations.

Results: E_a/E_b scalar fields

- Qualitative comparison between the fields of different populations
- Significant difference between the populations
- E_a/E_b is related to the flexibility of the axons, thus:
 - Normal axons have a more constant direction throughout its length, followed by mutant type 2 and mutant type 1

Conclusions

- 2D discrete stochastic model for the simulation of axonal biogenesis
- Several independent processes (elongation, shape and bifurcation) are considered
- Parameters estimated from real static data
- Validated on 53 real confocal microscopy images of single neurons (*Drosophila*)
- Generated synthetic axonal trees resemble real ones
- Parameters provide additional information on the populations

Future Work

- Improve validation
 - Better comparison between real/synthetic axons
 - Validation of the field
- Extend to 3D (or 4D)
- Include additional biological processes (speed, retraction, etc)

Thanks

Thank you!