Temporal adaptation of vascular patterning #40

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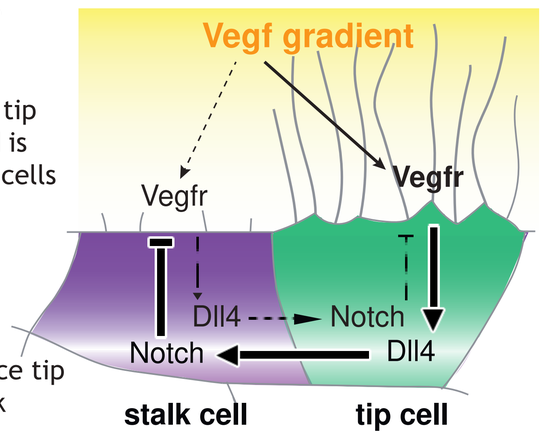
INTRODUCTION

Sprouting angiogenesis is highly dependent on effective decision making between endothelial cells (ECs). The Feedback between Vegf/Dll4/Notch is well established during the collective decision process selecting leading tip cells of new vessel sprouts. The tip cell selection speed is influencing the vasular network density. The faster tip cells are selected, the denser the network will bel.

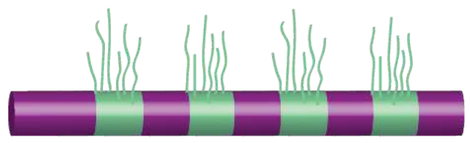
We recently showed that filopodia can act as temporal regulators and can increase the tip cell selection rate (Zakirov B. et al. R Soc PTB. 2021). We are currently investigating the mechanism by which filopodia influence tip cell selection rates and how this influences the network density. Additionally, we are aiming to uncover further temporal regulators of tip cell selection.



Our work will offer new targets for therapeutic approaches targeting temporal regulation of vascular network topology and branching density.

RESEARCH AIM

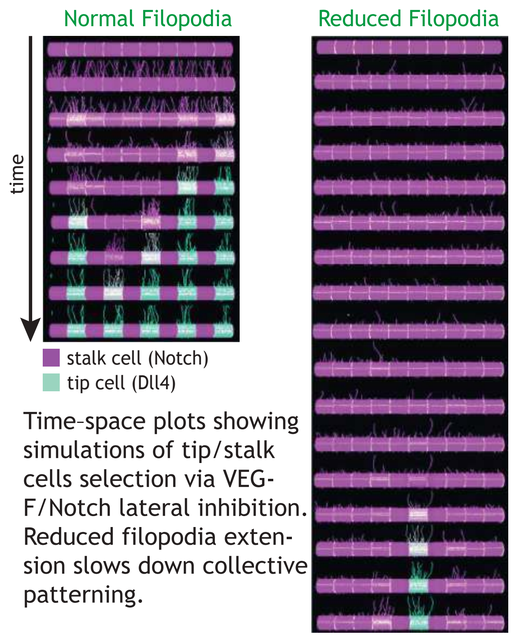
**I am developing a minimalistic model system to study tip cell selection using microfabrication.
This system will allow the dynamic observation of the selection process. It will serve as a testbed for temporal regulators of tip cell selection, as it allows for easy genetic and molecular manipulation.**



BACKGROUND

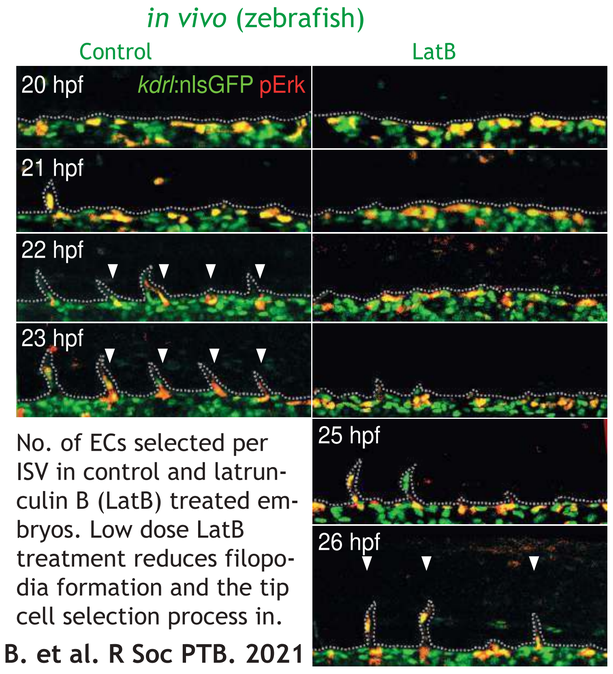
Loss of Filopodia reduces tip cells selection speed

in silico



Time-space plots showing simulations of tip/stalk cells selection via VEGF/Notch lateral inhibition. Reduced filopodia extension slows down collective patterning.

in vivo (zebrafish)

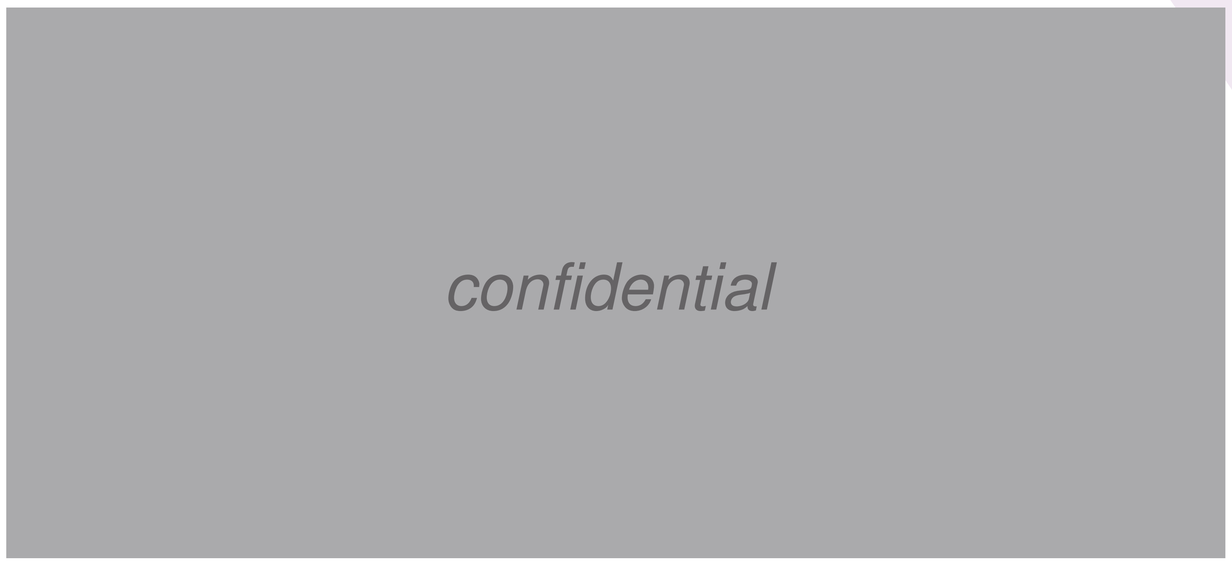


No. of ECs selected per ISV in control and latrunculin B (LatB) treated embryos. Low dose LatB treatment reduces filopodia formation and the tip cell selection process in.

Zakirov B. et al. R Soc PTB. 2021

APPROACH Device 1

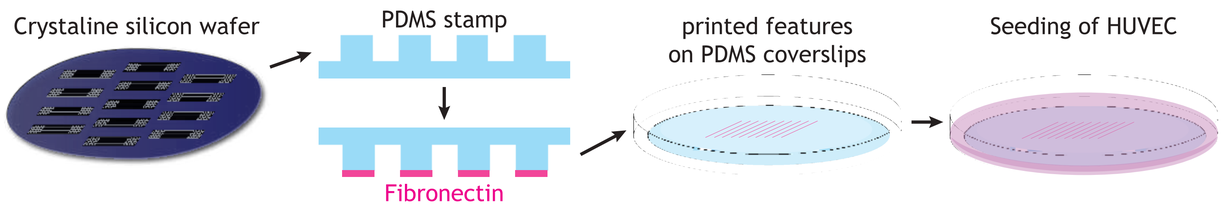
PDMS channel chip



PDMS (Polydimethylsiloxane) channel chips are made using crystaline silicon wafers. The channel chip is bonded to PDMS coated coverslips and coated with fibronectin. Cells are flushed in using needles and the gradient reservoir is filled with medium containing VEGFa.

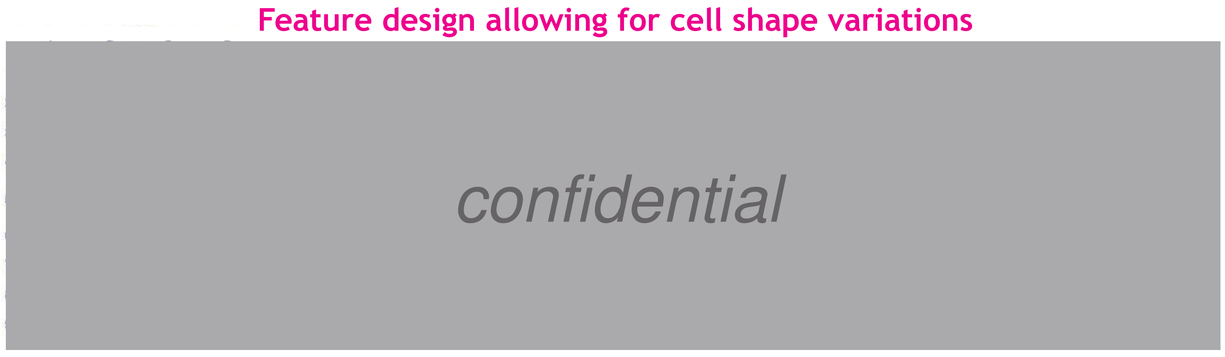
APPROACH Device 2

Feature preparation using PDMS stamps

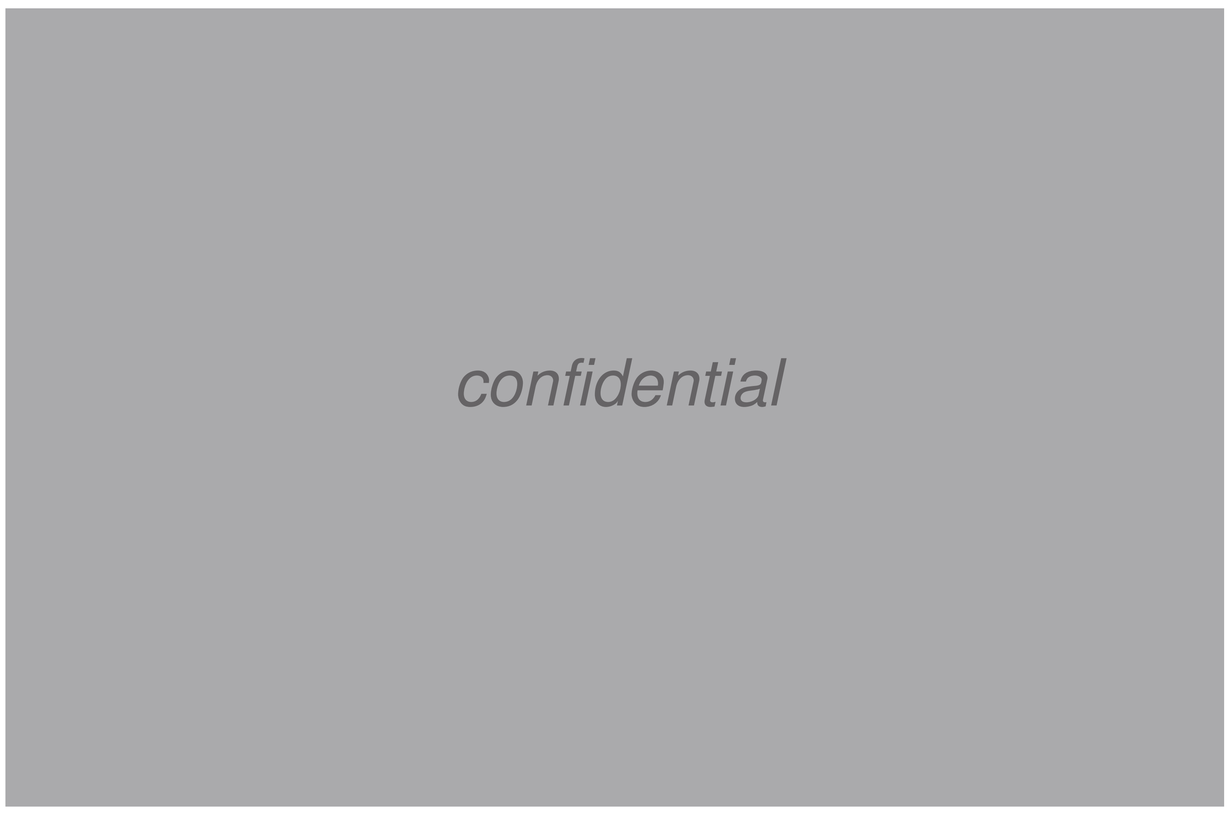


PDMS (Polydimethylsiloxane) stamps are made using crystaline silicon wafers. The stamps are incubated with fibronectin which is printed onto PDMS coated coverslips. Areas outside the features are covered in F127 to stop cells from adhering. Subsequently cells are seeded on the features.

Feature design allowing for cell shape variations

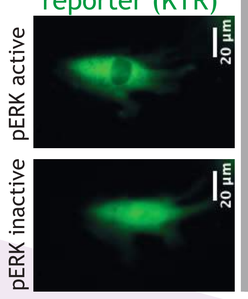


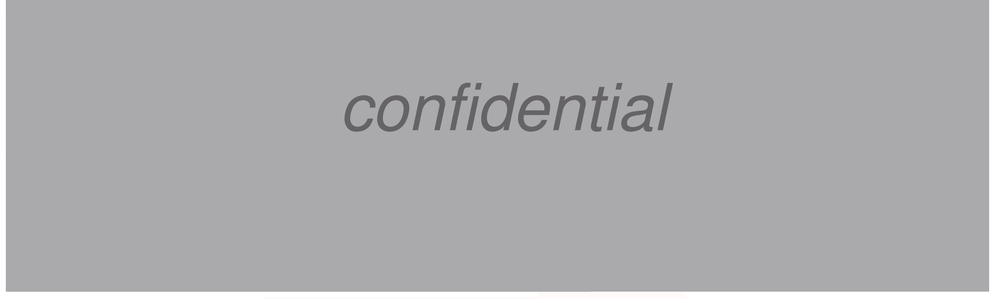
RESULTS Device 2



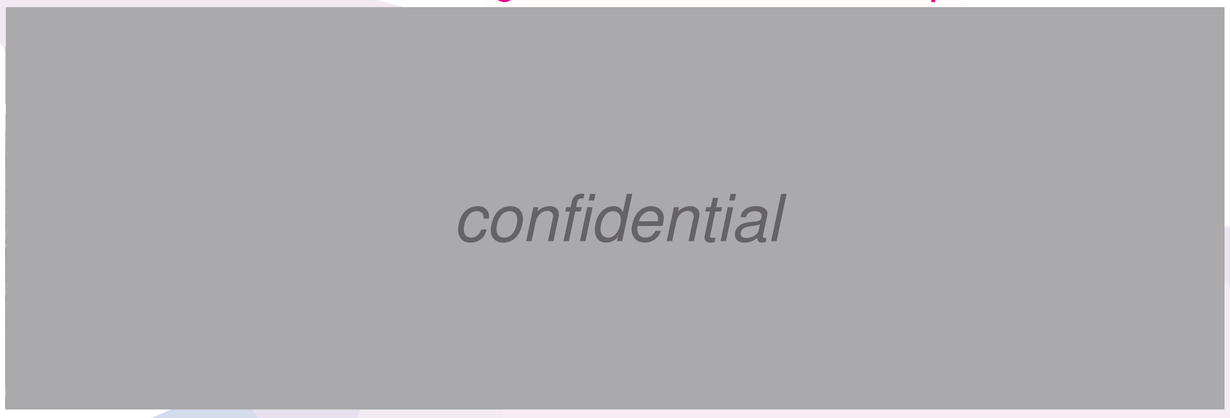
pERK as dynamic tip cell marker

ERK-kinase translocation reporter (KTR)





HUVEC show Notch regulated differential ERK-KTR patterns

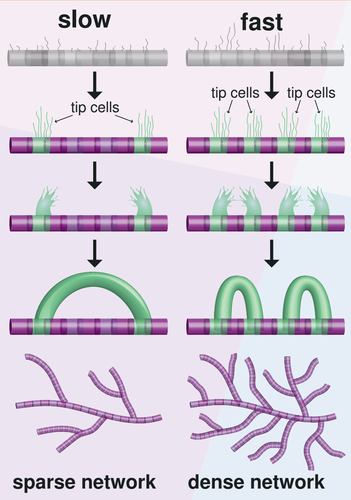


CONCLUSION and OUTLOOK

Our prior work demonstrates that altered tip selection speed can modulate branching density and overall network topology. Thus, we aim to better understand how the temporal regulation of tip cell selection is achieved during sprouting angiogenesis. For example the role of filopodia as temporal regulators.

Using microcontact printing and microfabrication I am developing a minimalist model system, allowing to closely monitor and control the tip/stalk cell selection while modulating the involvement of specific cellular components.

This model system will serve as testbed to identify additional temporal regulators of tip cell selection.



FUNDING AND REFERENCES

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