An integrated computational approach for the design of patient-specific virtual tumours

Angélique Stéphanou

Laboratoire TIMC-IMAG / DyCTiM

Context

Important progress has been made in cancer therapy over the past 10 years

- a wider range of therapeutic options
- more specific molecules that reduce side effects
- more individualized therapies

Context

Important progress has been made in cancer therapy over the past 10 years

- a wider range of therapeutic options
- more specific molecules that reduce side effects
- more individualized therapies

However, tumour development becomes more complex with therapies

- resistance to treatment
- selection of the most aggressive phenotypes

Context

Important progress has been made in cancer therapy over the past 10 years

- a wider range of therapeutic options
- more specific molecules that reduce side effects
- more individualized therapies

However, tumour development becomes more complex with therapies

- resistance to treatment
- selection of the most aggressive phenotypes

The new objectives

- rationalize the use of the therapies
- combine the therapies to enhance the effects
- adapt the therapies with the evolution of the tumour and patient states

Why the need for a virtual tumour ?

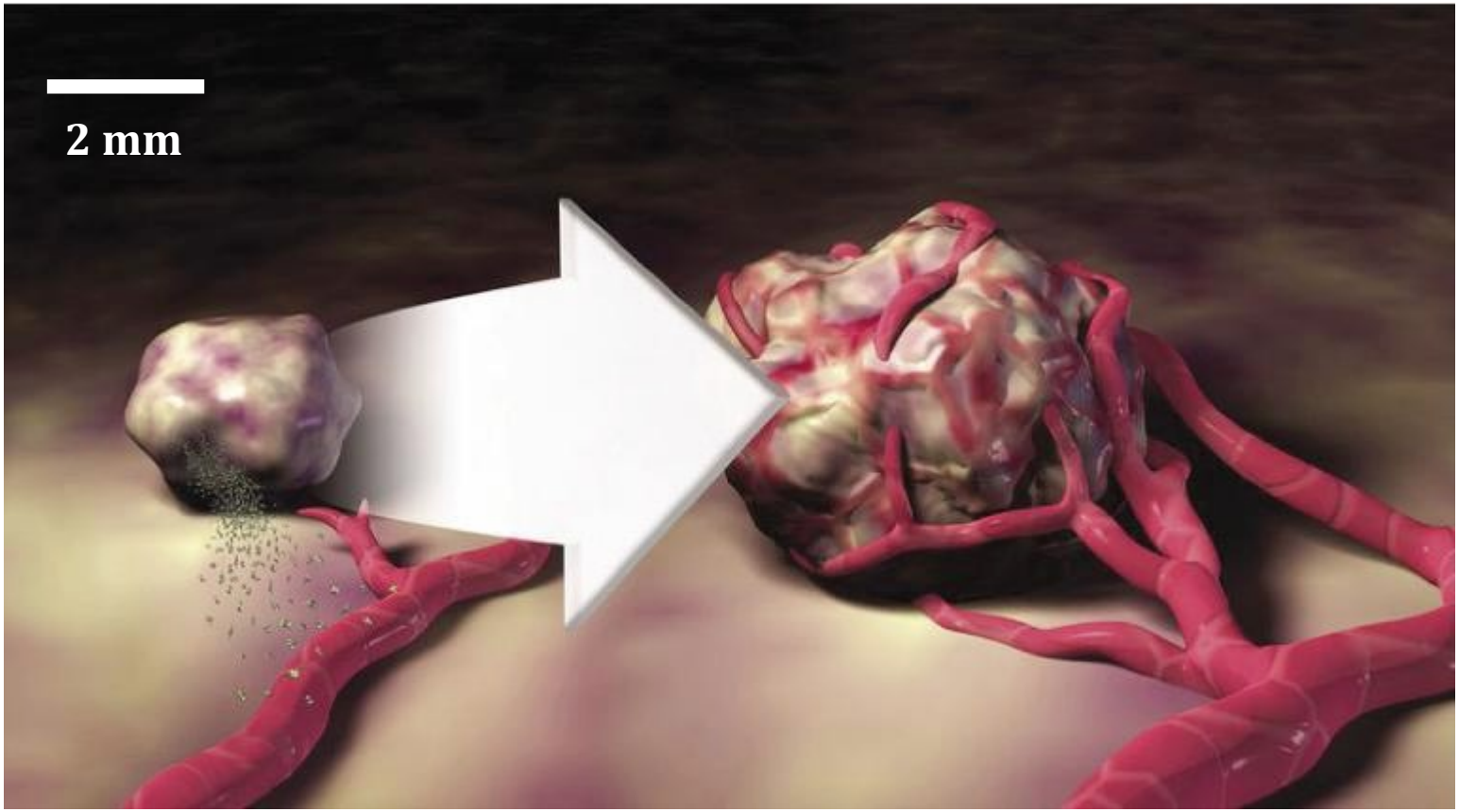
Provide an integrated vision of the phenomenon

Gives the ability to make virtual experiments

- to test hypotheses
- to better understand the system
- to predict the system's reaction
- to optimize the solutions

Development of a solid tumour

Avascular growth **Vascular growth**



hypoxia induces VEGF release

VEGF induces angiogenesis

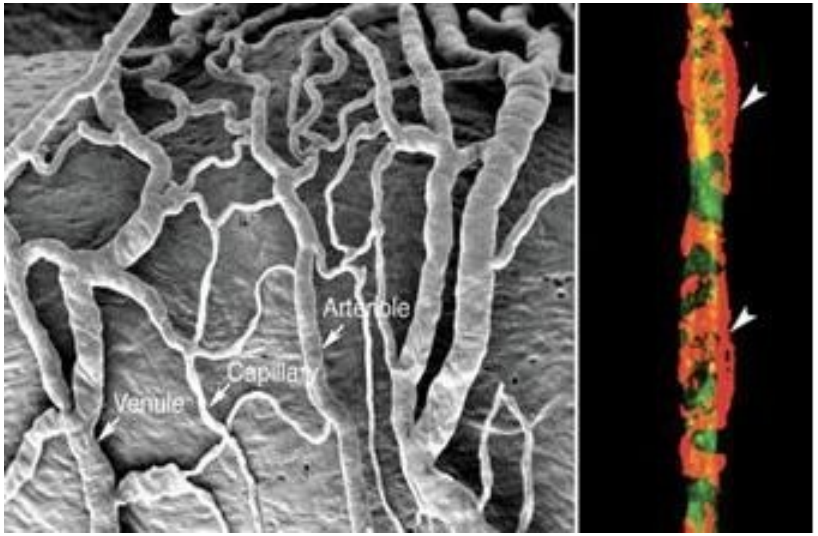
Development of a solid tumour

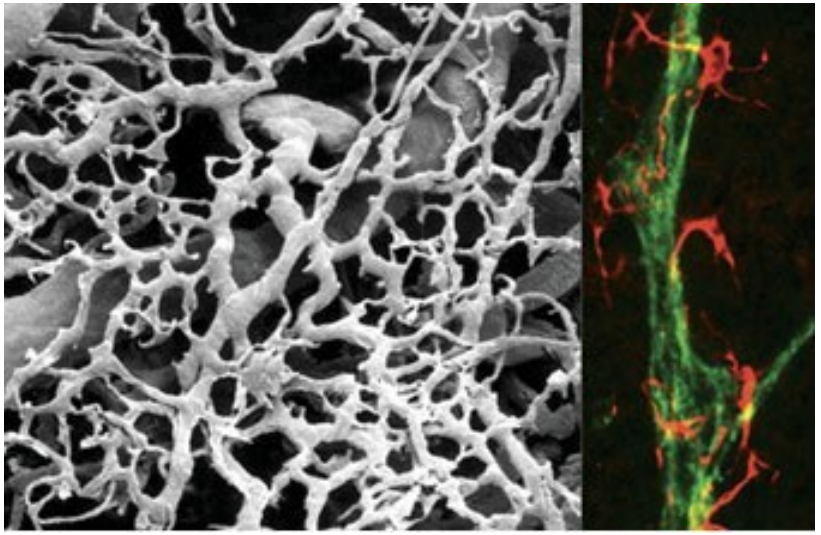
Avascular growth **Vascular growth**

hypoxia induces VEGF release

Cells escape from the tumour mass

Normal *vs* tumoral vascular network





Normal network

- organized network
- impermeable vessels
- pericytes coverage (red)

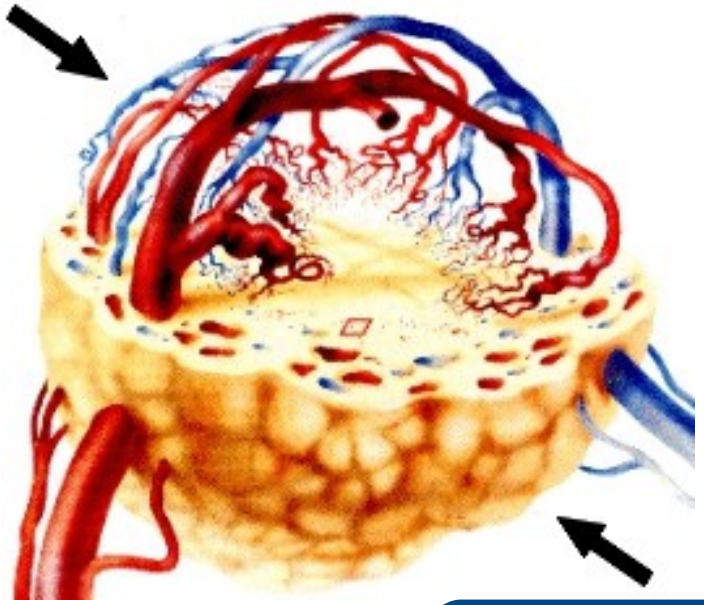
Tumour network

- disorganized (abnormal, dense and tortuous)
- permeable vessels
- no pericytes coverage (red)

Therapeutic modes under consideration

Vascular disrupting agents

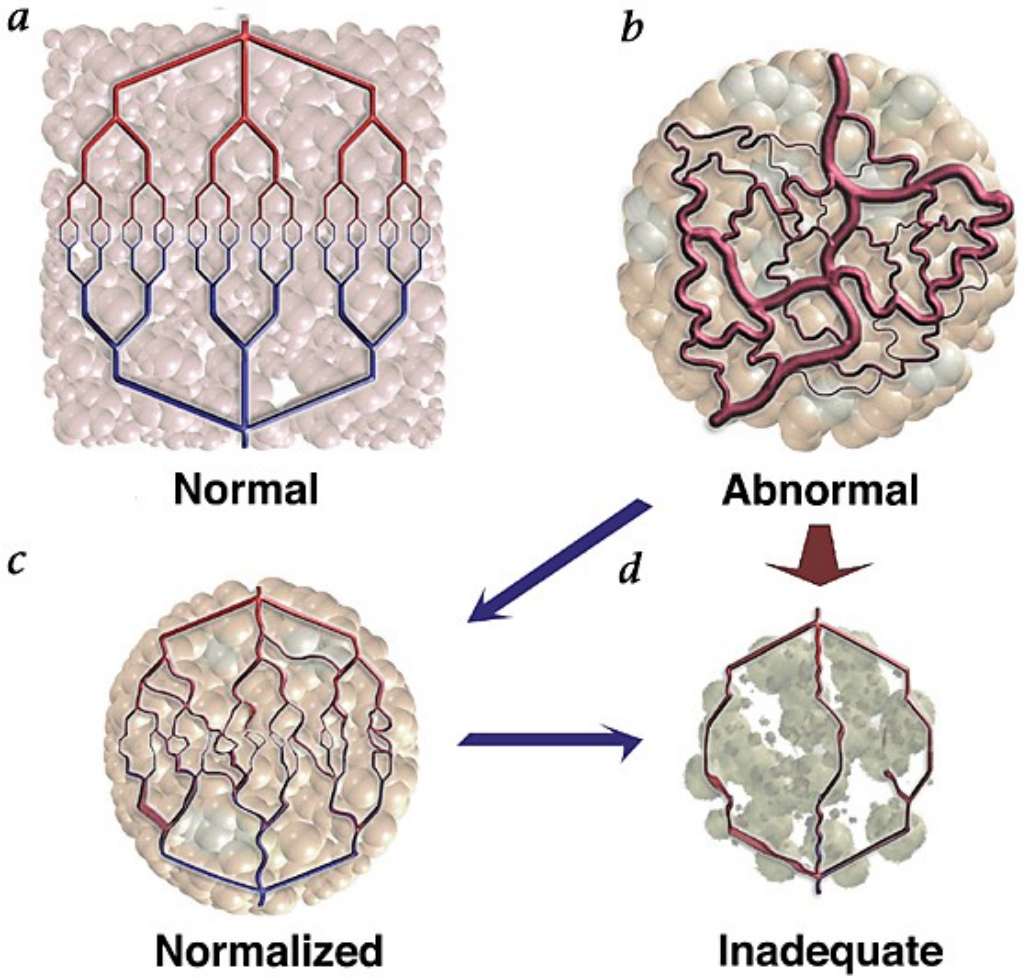
target = vessels



Cytotoxic molecules

target = tumour cells

Therapeutic coupling

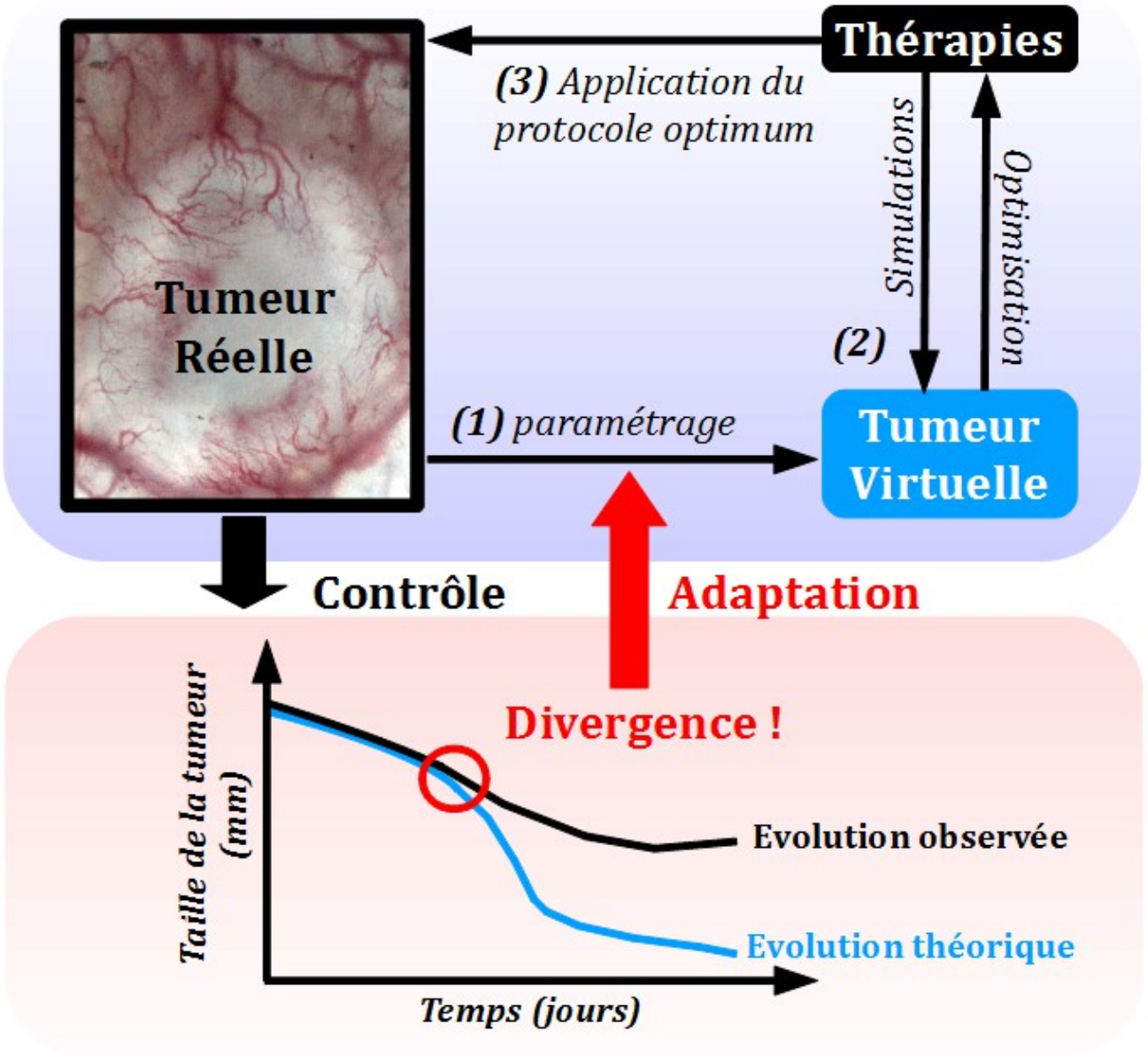


VDA can **temporarily normalize** the tumour vascular network

The delivery of cytotoxic molecules to the tumour is **transiently improved**

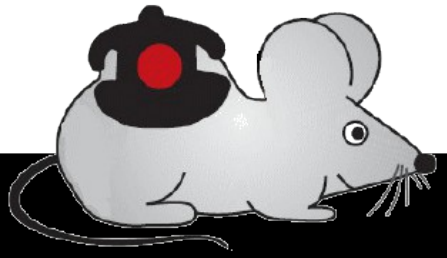
Therapeutic window

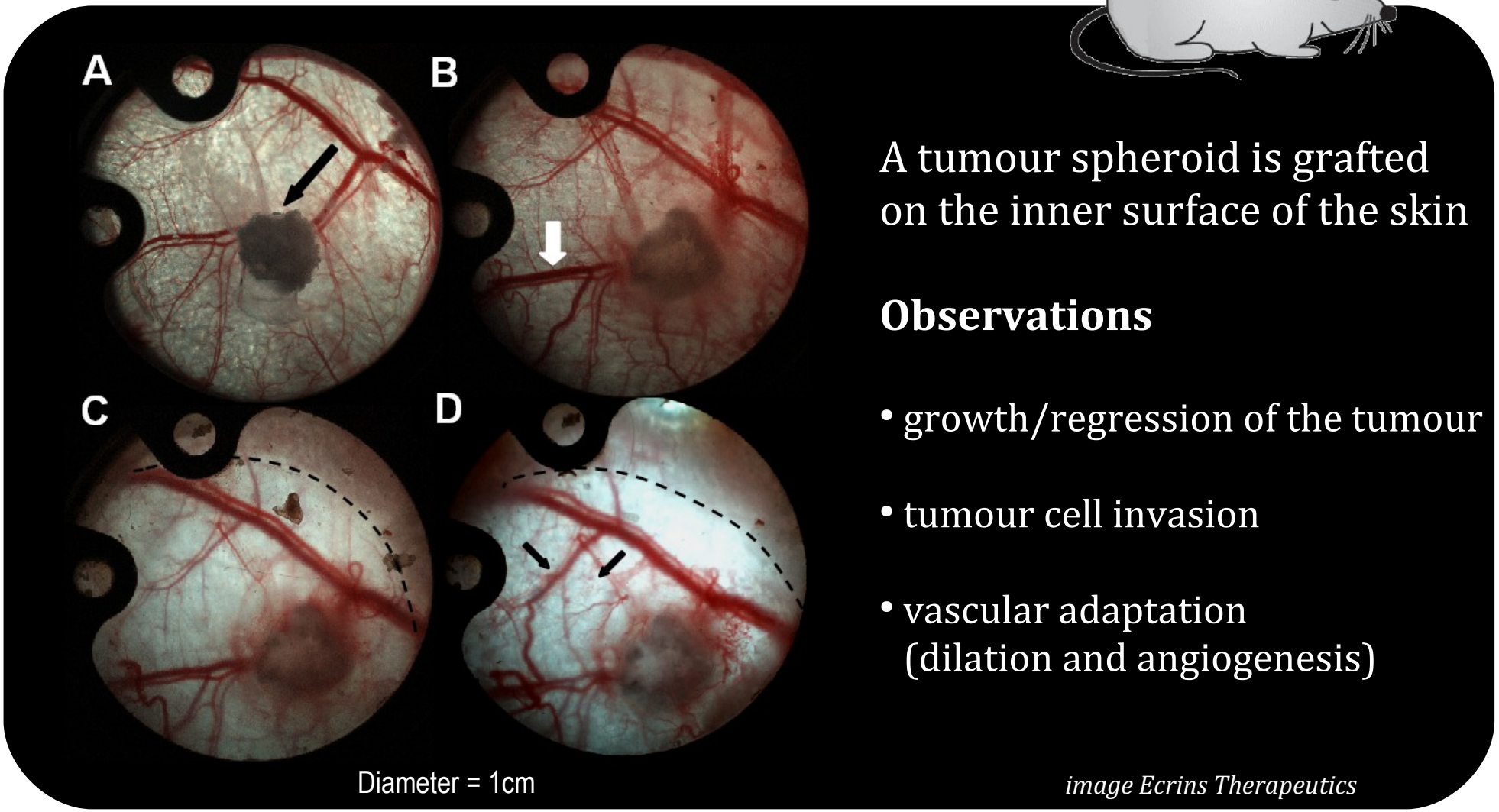
Computer-assisted therapeutic strategy



The experimental model (1)

in vivo observations with a dorsal chamber





A tumour spheroid is grafted on the inner surface of the skin

Observations

- growth/regression of the tumour
- tumour cell invasion
- vascular adaptation (dilation and angiogenesis)

image Ecrins Therapeutics

Vascular adaptation

Day 1

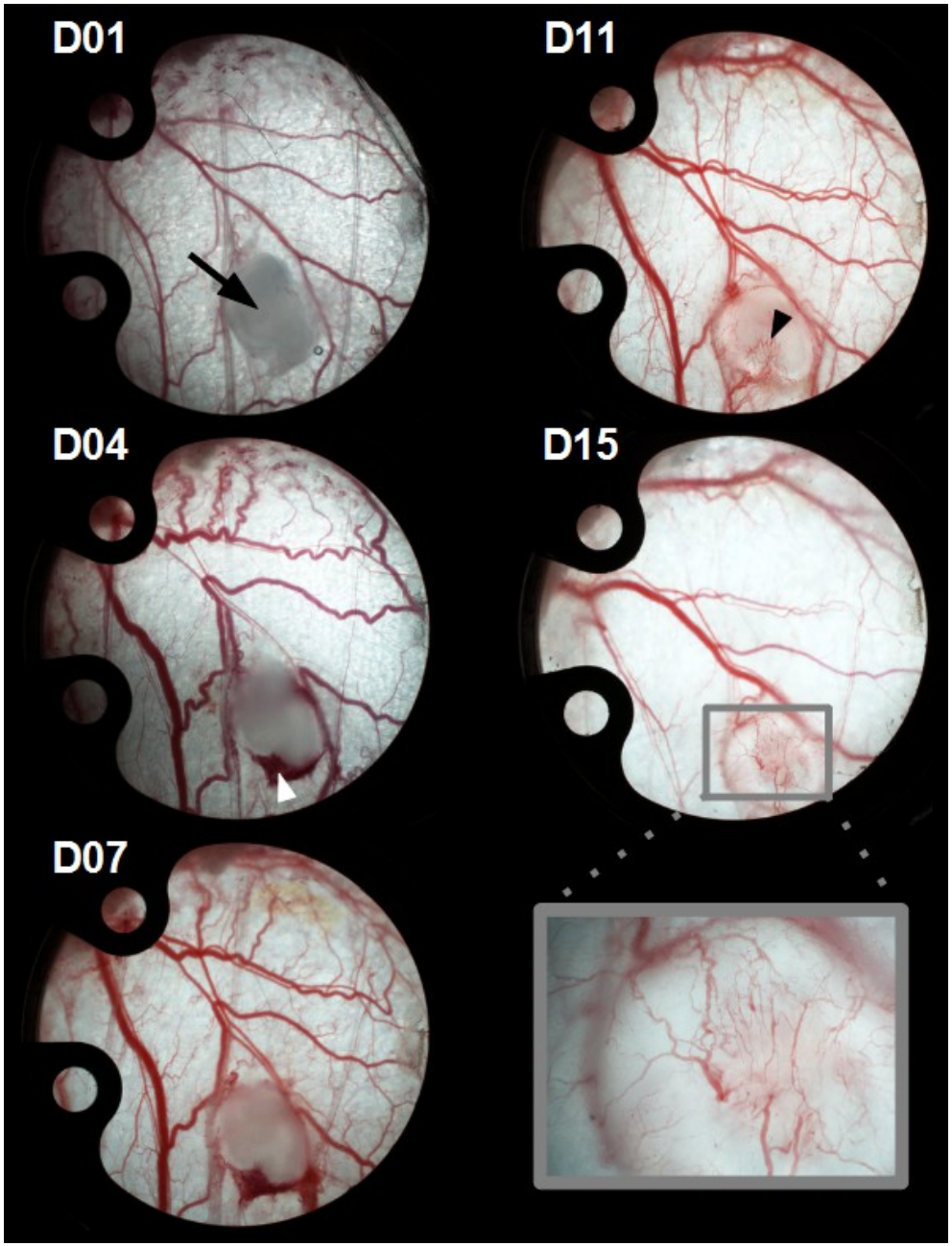
Implantation of the tumour spheroid

Day 4 to day 7

Vessels dilation and haemorrhage induced by the growth factors (VEGF)

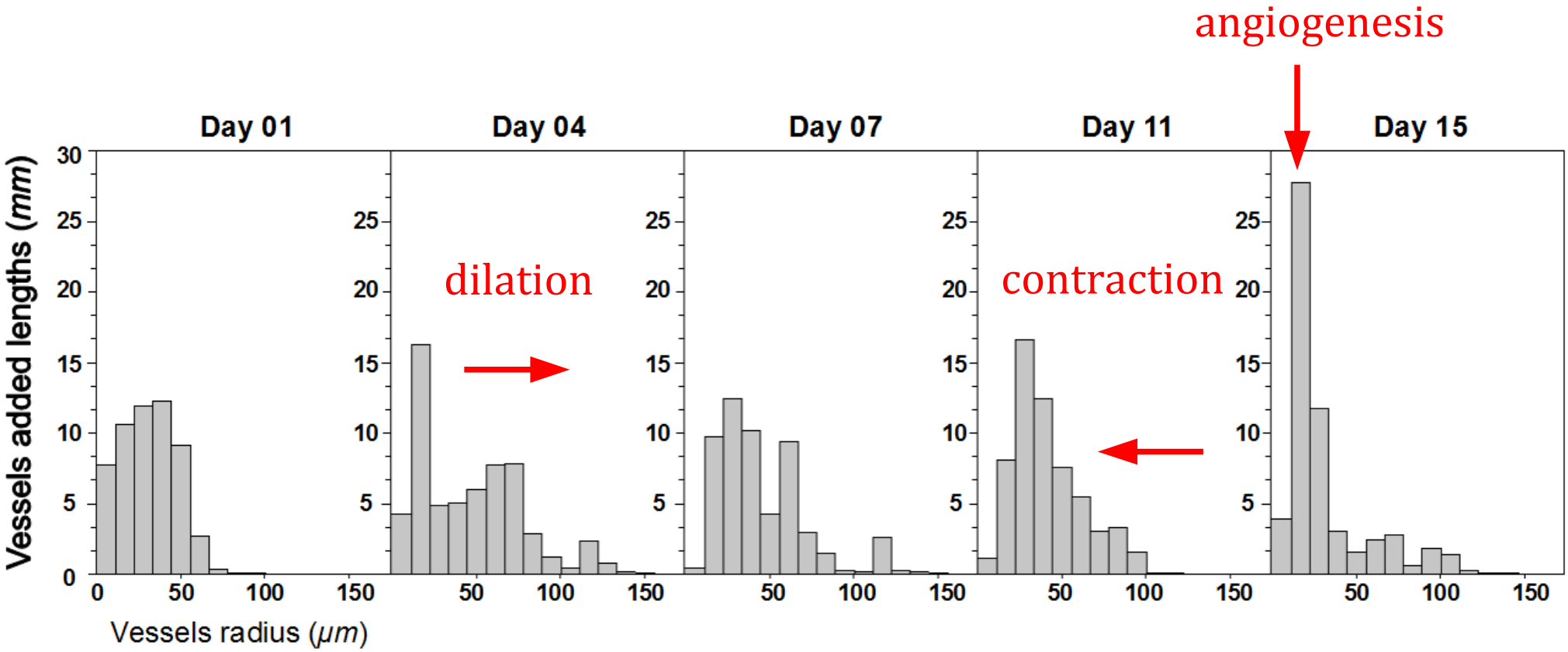
Day 11 to day 15

Contraction of the vessels and angiogenesis



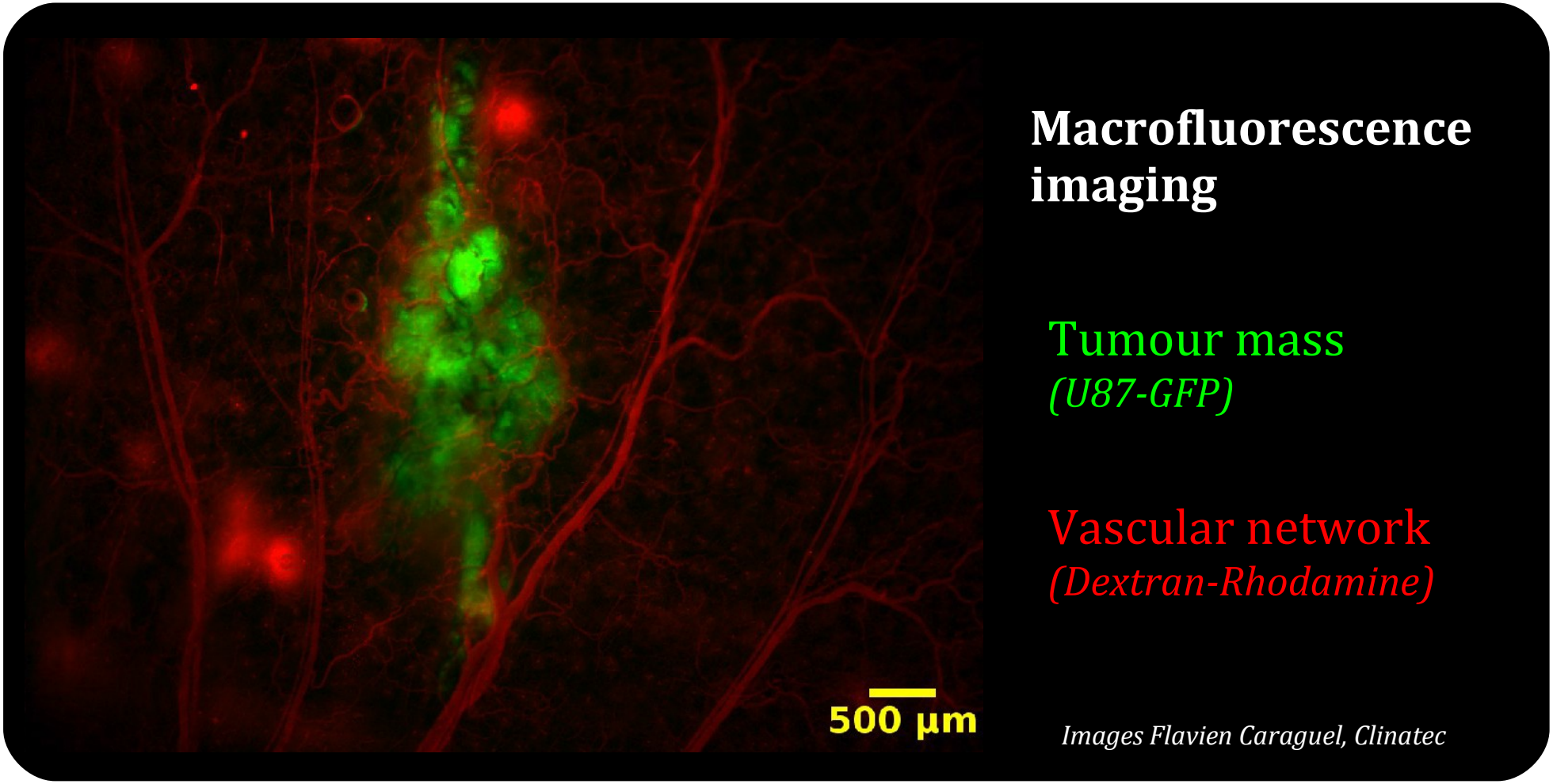
Vascular adaptation

Evolution of the cumulated length of the vessels in each range of vessels radii [0,10], [10,20] ... [140,150] µm



The experimental model (2)

in vivo observation through the mouse pinna



Macrofluorescence imaging

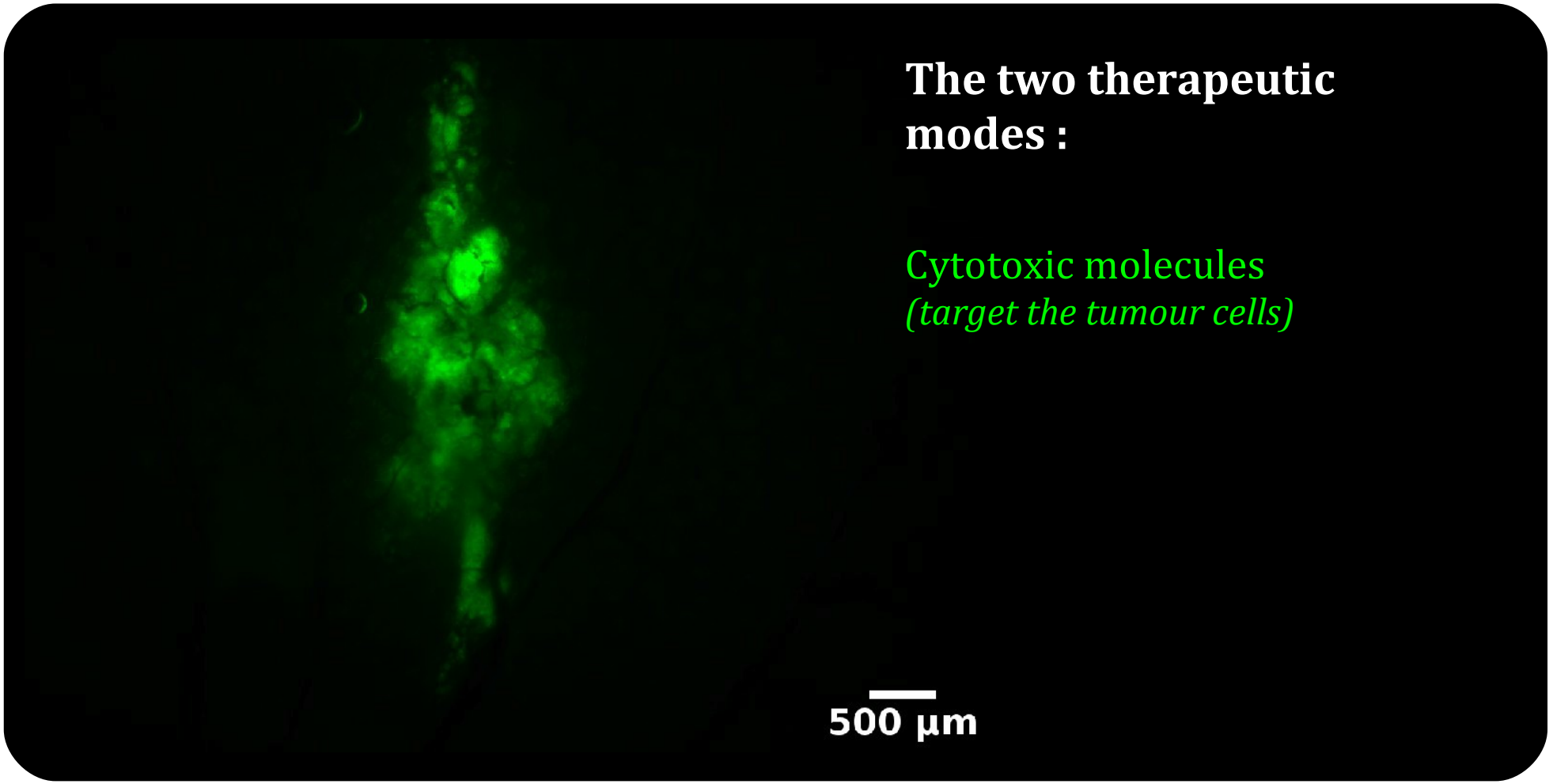
Tumour mass
(U87-GFP)

Vascular network
(Dextran-Rhodamine)

Images Flavien Caraguel, Clinatec

The experimental model (2)

in vivo observation through the mouse pinna

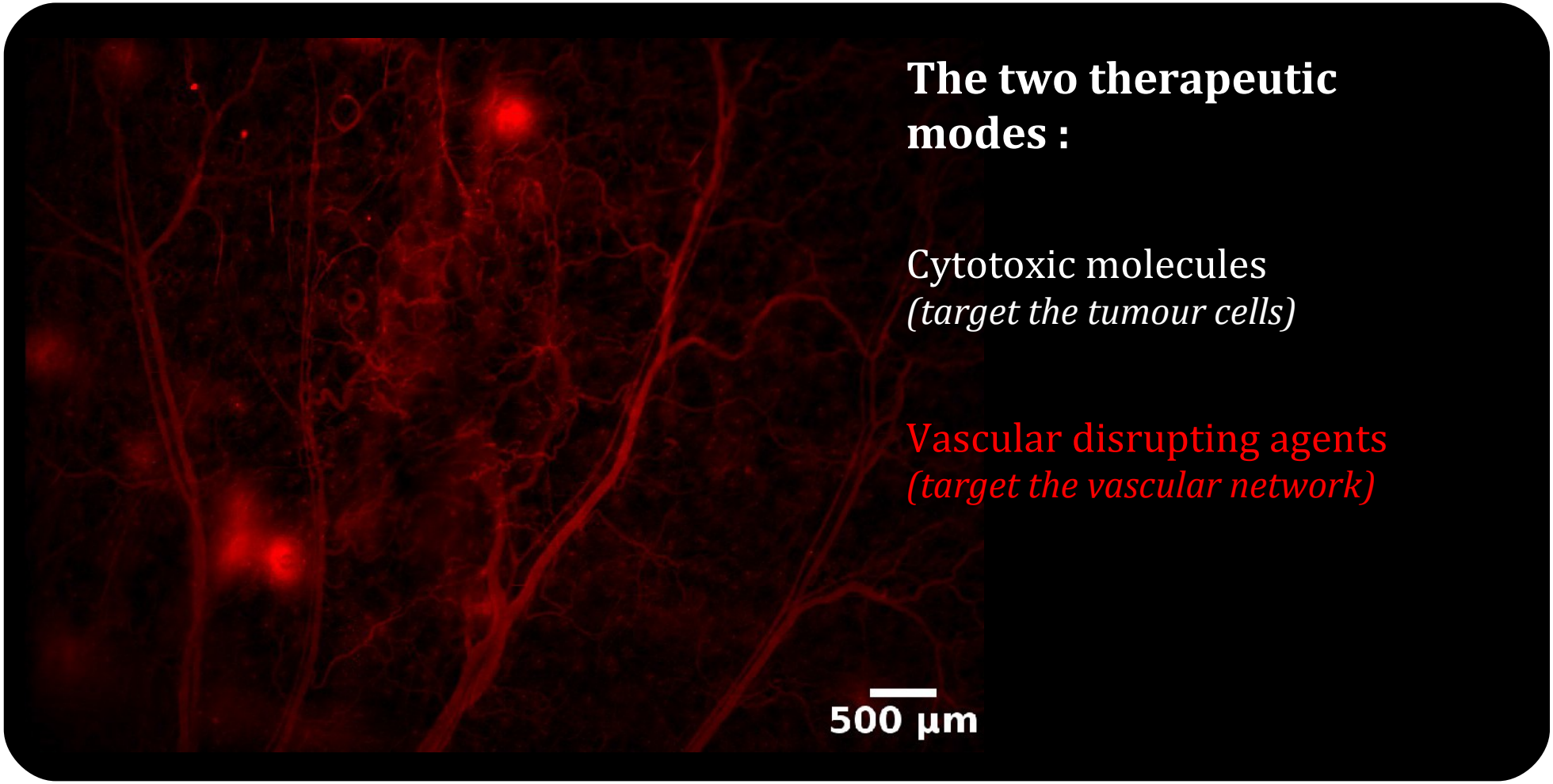


The two therapeutic modes :

Cytotoxic molecules
(target the tumour cells)

The experimental model (2)

in vivo observation through the mouse pinna



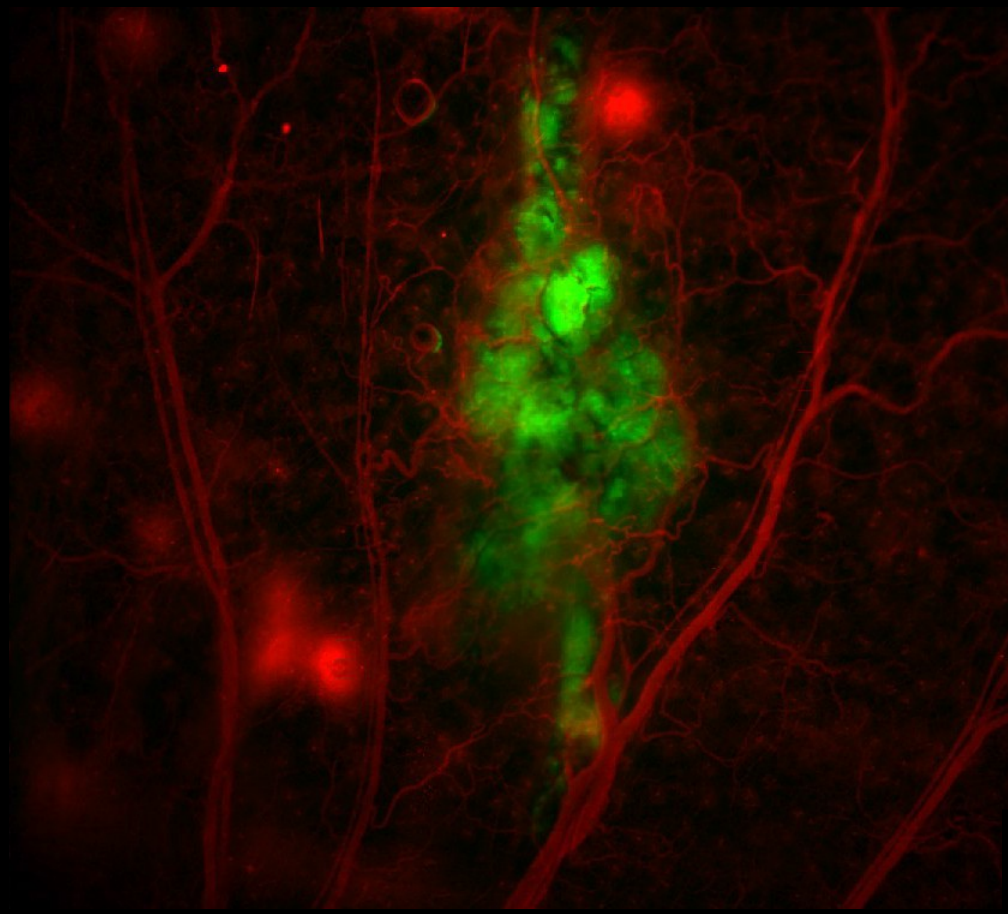
The two therapeutic modes :

Cytotoxic molecules
(target the tumour cells)

Vascular disrupting agents
(target the vascular network)

The experimental model (2)

in vivo observation through the mouse pinna



The two therapeutic modes :

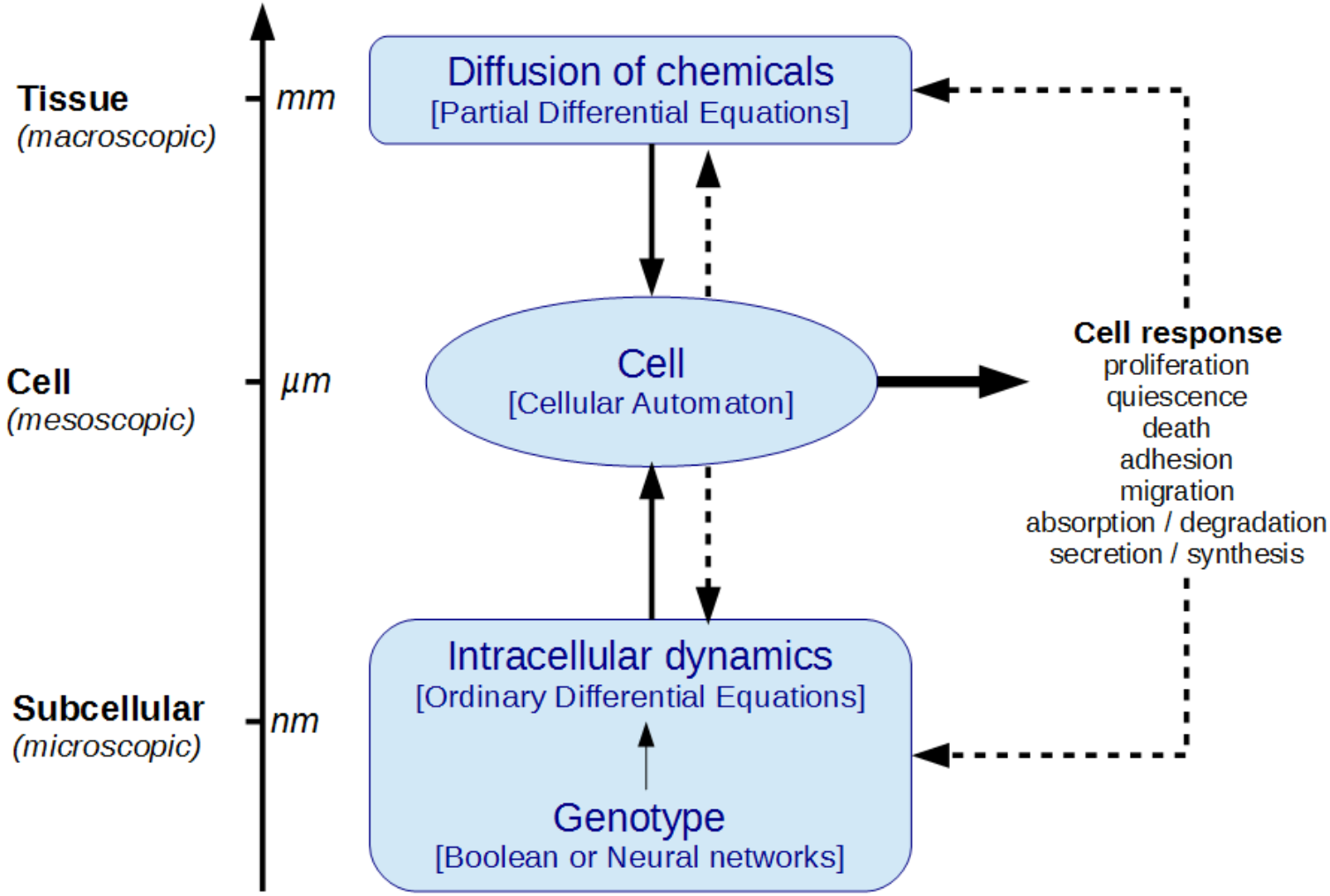
Cytotoxic molecules
(target the tumour cells)

+

Vascular disrupting agents
(target the vascular network)

Research of synergetic effects

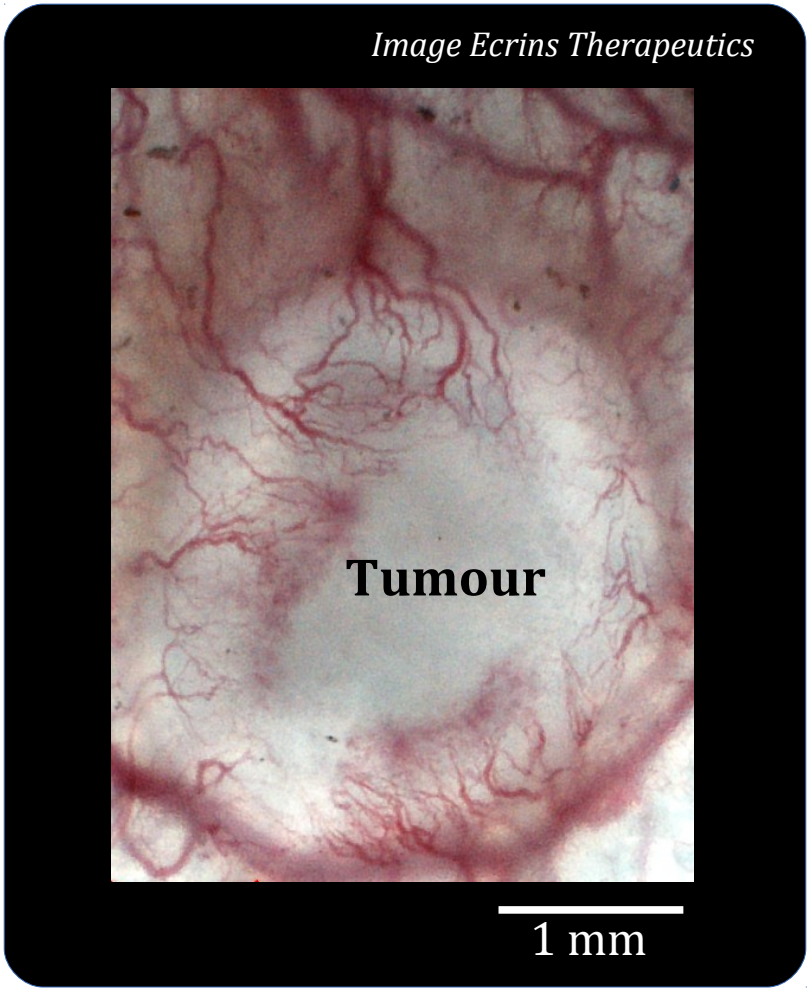
A cell-centred hybrid multiscale model



The computational model

Tumour growth, metastatic invasion
Cell cycle, metabolism and death

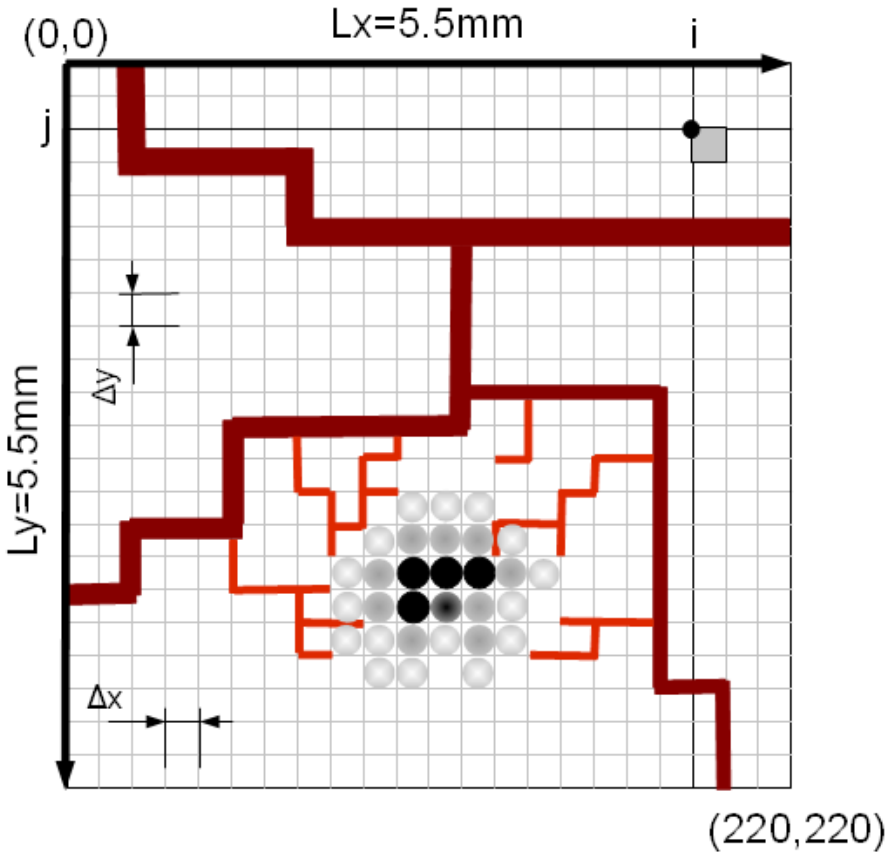
Angiogenesis, vascular adaptation
Vessels permeability, diffusion of therapeutic molecules and oxygen



The computational model

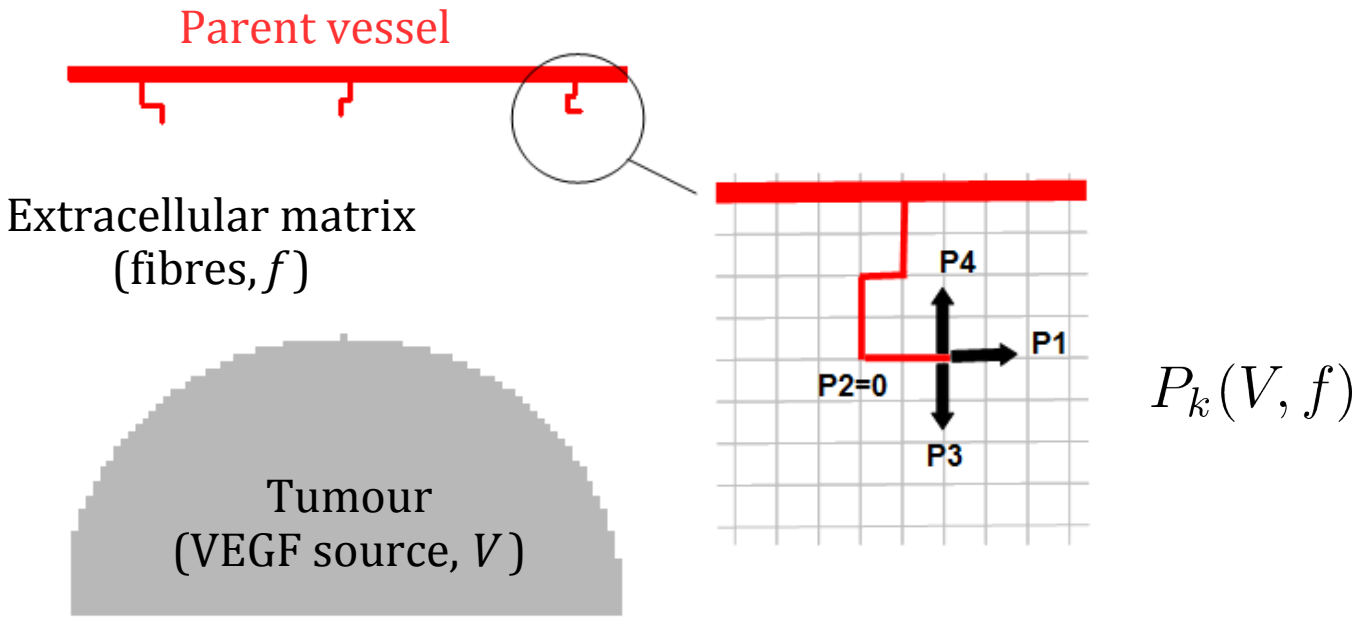
Tumour growth, metastatic invasion
Cell cycle, metabolism and death

Angiogenesis, vascular adaptation
Vessels permeability, diffusion of therapeutic molecules and oxygen



Angiogenesis modelling

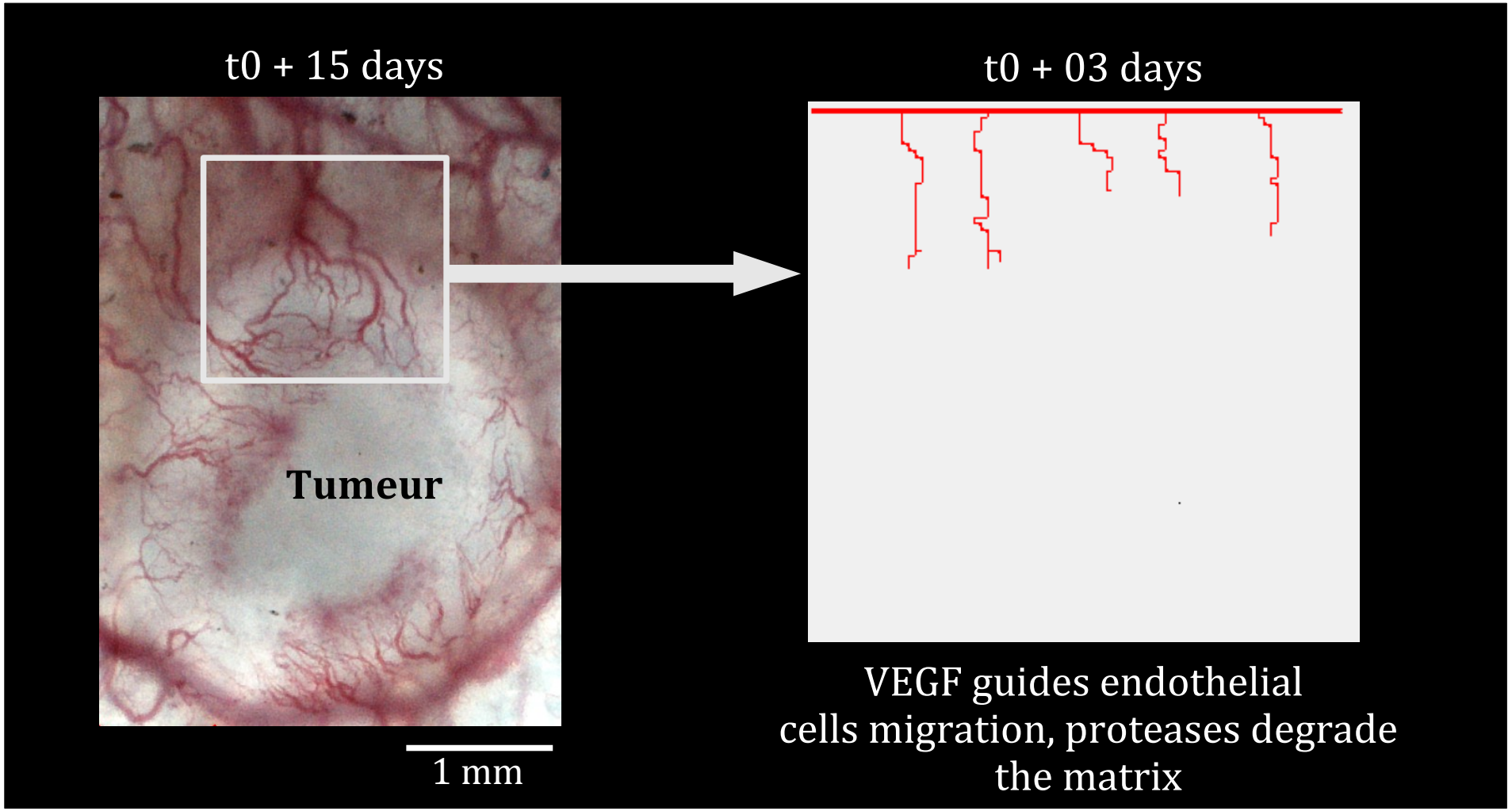
Tumour vascularization develops through the extracellular matrix *(f)* in response to growth factors *(V)* produced by the tumour cells in hypoxia because of cell overcrowding.



$$n_{i,j}^{t+1} = P_0 n_{i,j}^t + P_1 n_{i+1,j}^t + P_2 n_{i,j-1}^t + P_3 n_{i,j+1}^t + P_4 n_{i,j-1}^t$$

Anderson and Chaplain, *Bull. Math. Biol.*, 1998

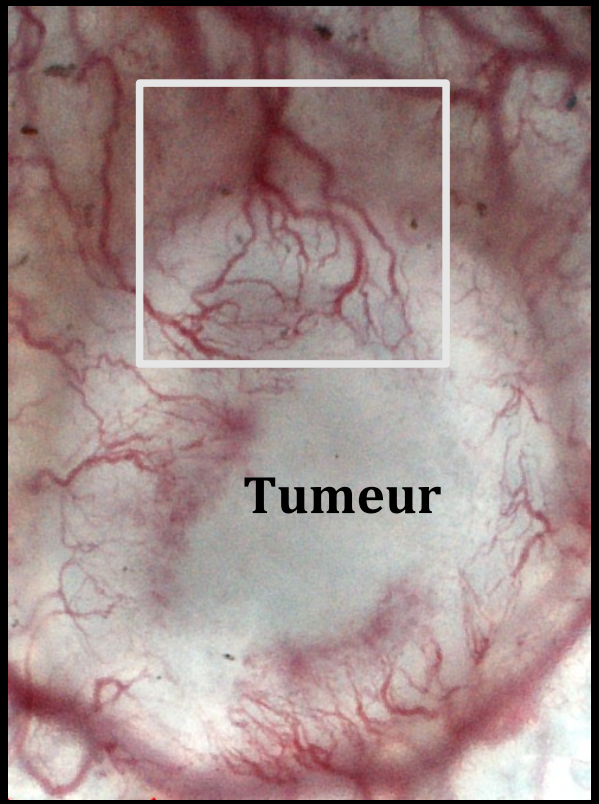
Simulation of angiogenesis



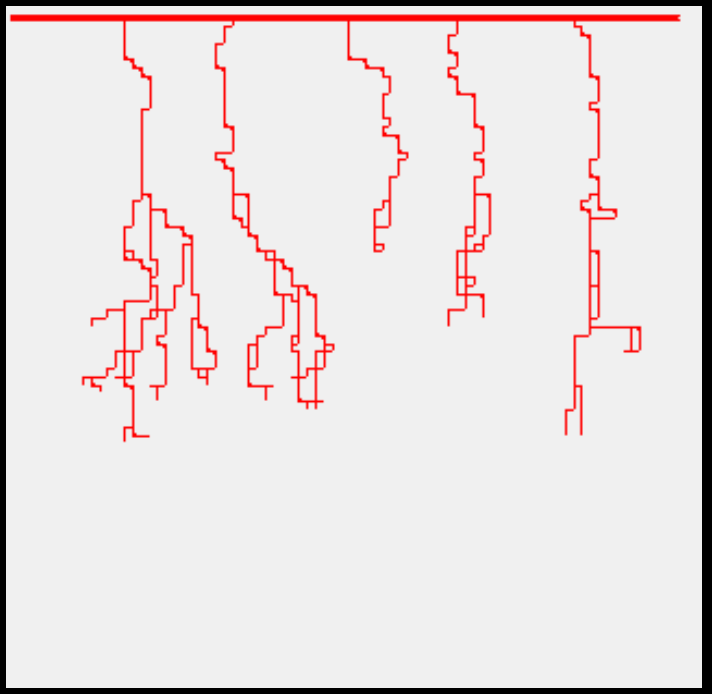
Stéphanou *et al.*, *Math Comp Mod,* 2005

Simulation of angiogenesis

t0 + 15 days



t0 + 08 days

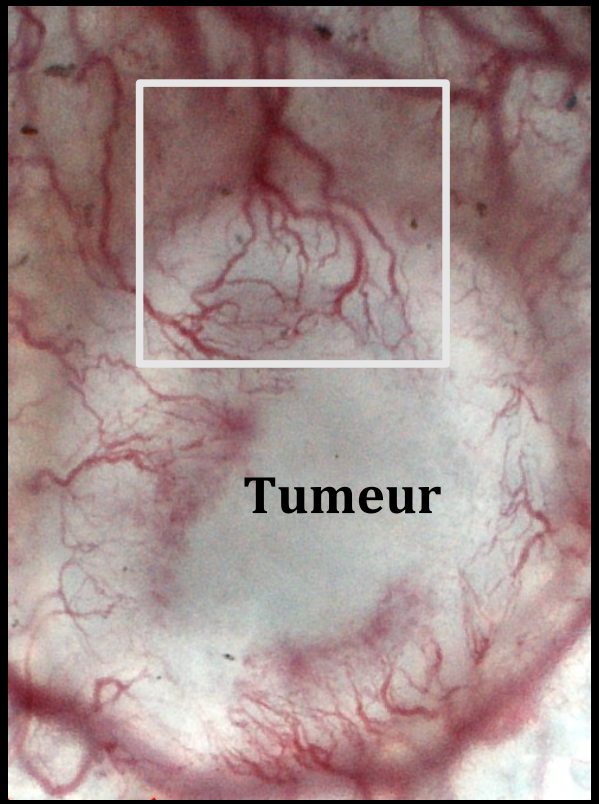


Sensitivity to the matrix mechanical properties

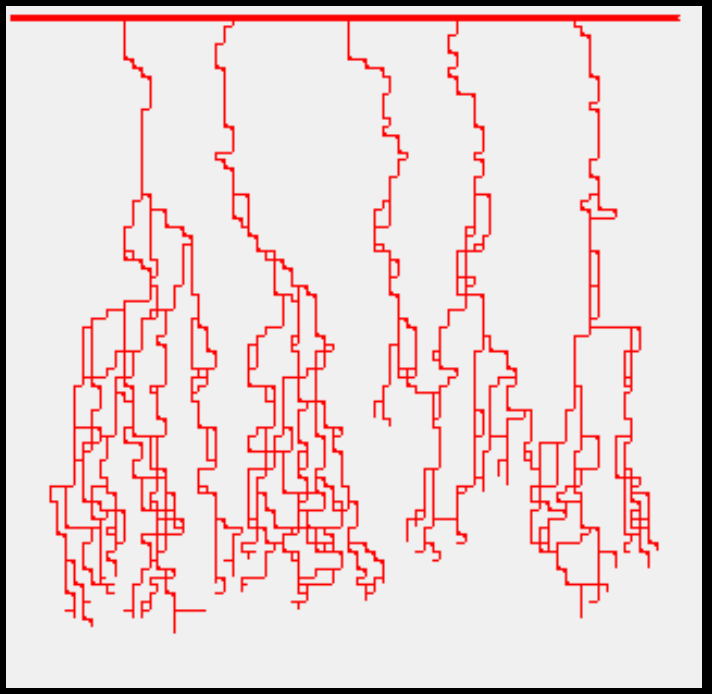
Stéphanou *et al.*, *Math Mod Nat Phenom,* 2015

Simulation of angiogenesis

t0 + 15 days



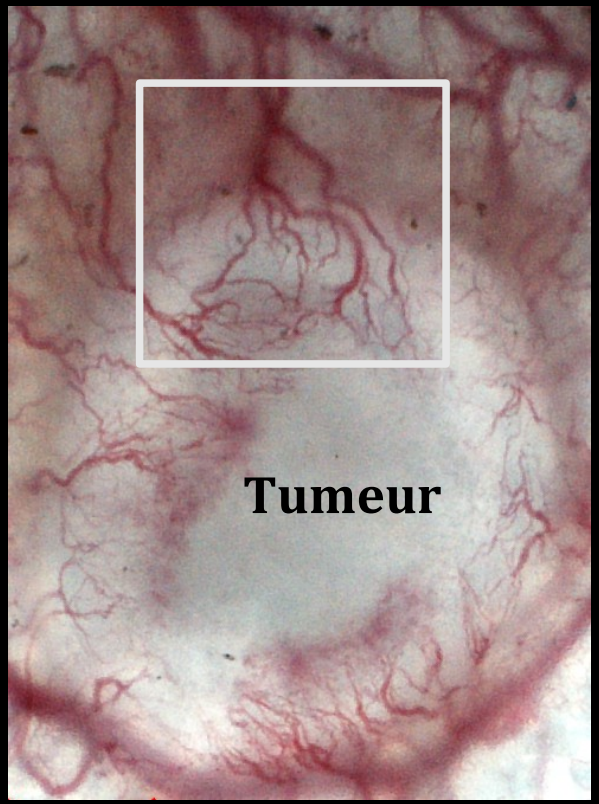
t0 + 12 days



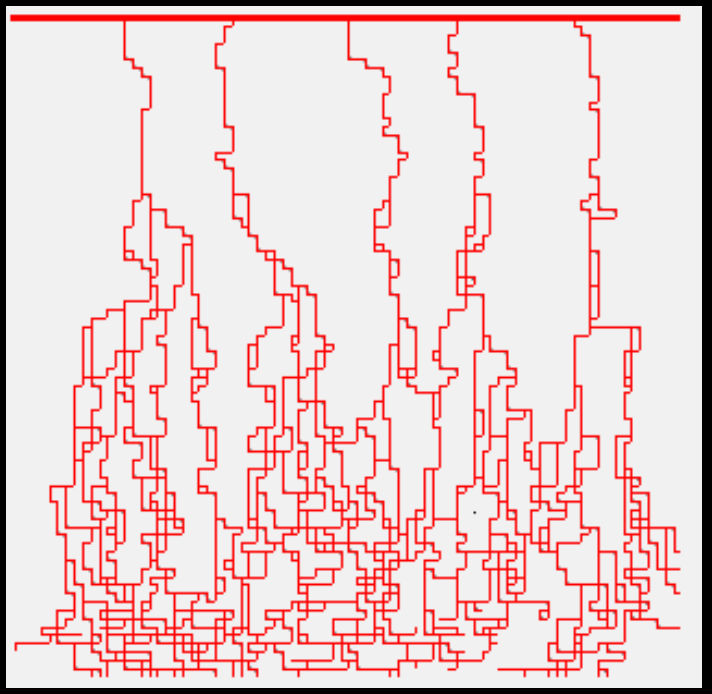
VEGF stimulates cell proliferation

Simulation of angiogenesis

t0 + 15 days



t0 + 15 days

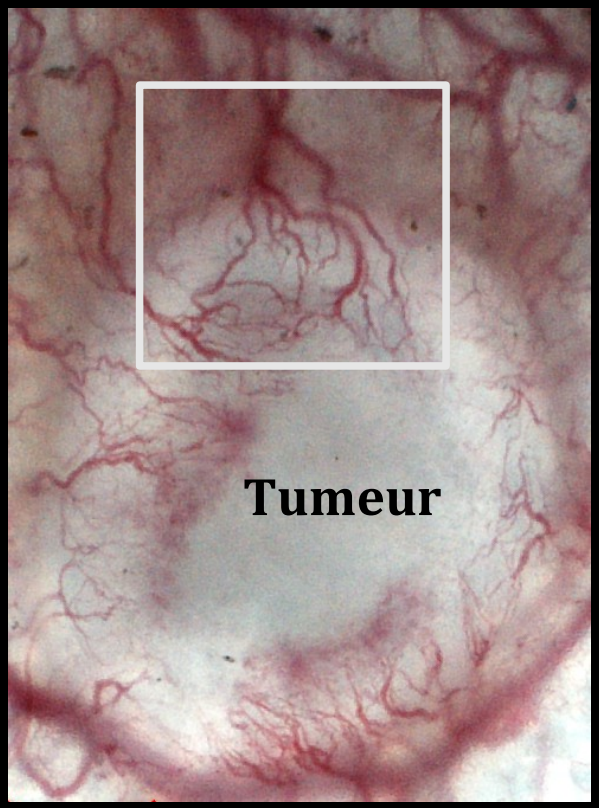


Necessity to functionalized the vascular network

Stéphanou *et al.*, *Math Comp Mod,* 2006

Simulation of angiogenesis

t0 + 15 days

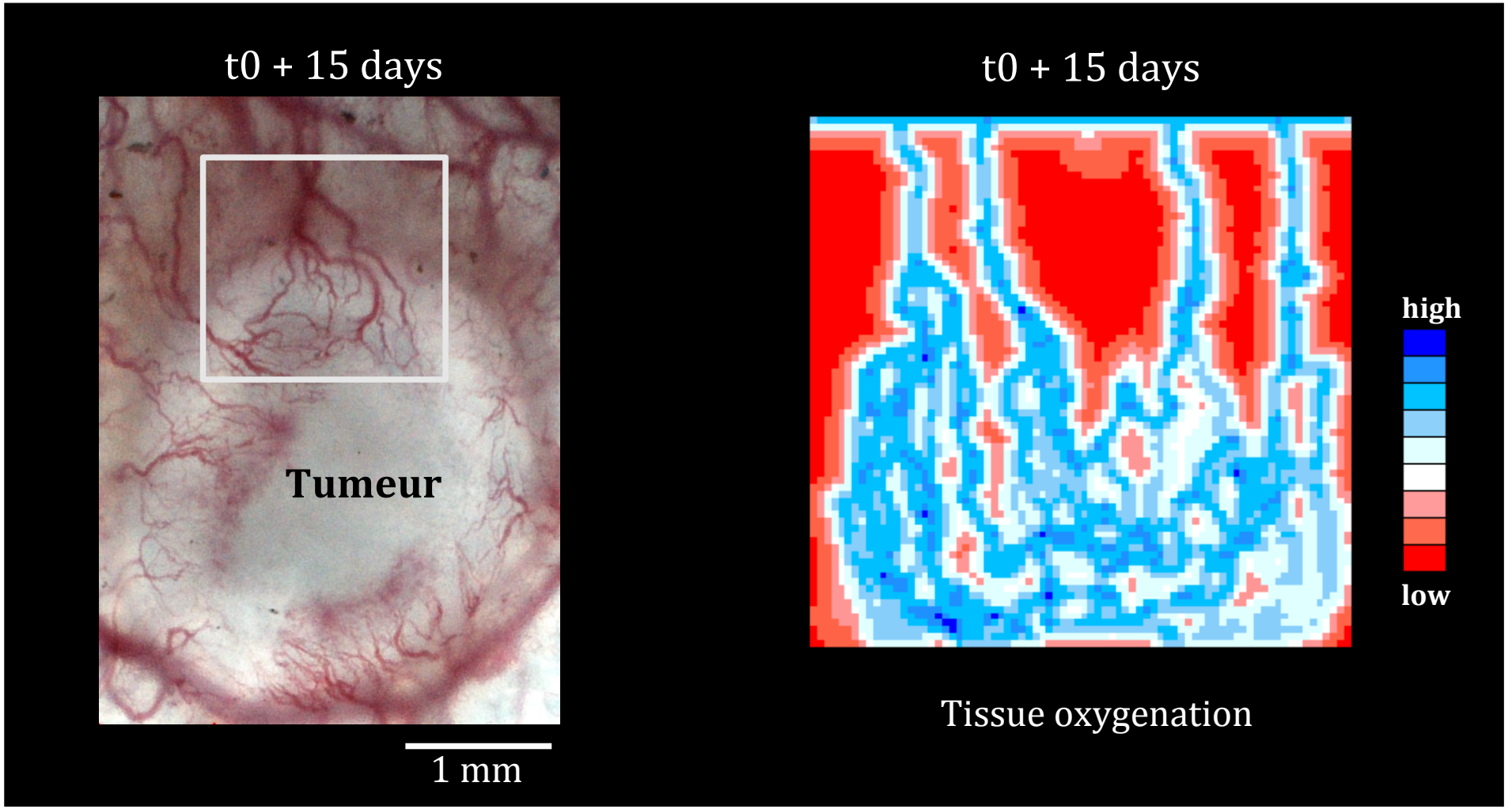


t0 + 15 days

Adaptation to hemodynamical constraints (blood flow)

$$\Delta R = \underbrace{\log(\tau_w + \tau_{ref})}_{\text{shear stress}} - \underbrace{\log(\tau_e P)}_{\text{transmural pressure}} + \underbrace{k_m \log\left(\frac{Q_{ref}}{Q H_D}\right)}_{\text{metabolic stimulus}} - \underbrace{k_s}_{\text{pericytes contractility}}$$

Simulation of angiogenesis



Pons-Salort *et al., Math Mod Nat Phenom,* 2012

Diffusive species

Proteases

Matrix degrading enzyme *(m)*

$$\frac{\partial m}{\partial t} = D_m \nabla^2 m + \alpha_m n_{i,j} - \nu_m m$$

Endothelial cell

Vascular degrading enzyme *(p)*

$$\frac{\partial p}{\partial t} = D_p \nabla^2 p + \alpha_p P_{i,j} - \nu_p p$$

Proliferative cell

Growth factors *(V)*

$$\frac{\partial V}{\partial t} = D_V \nabla^2 V + \alpha_V Q_{i,j} - \nu_V V - \lambda_V W_{i,j} min(V, V_{max})$$

Quiescent cell *Vessels "weight"*

Oxygen *(O)*

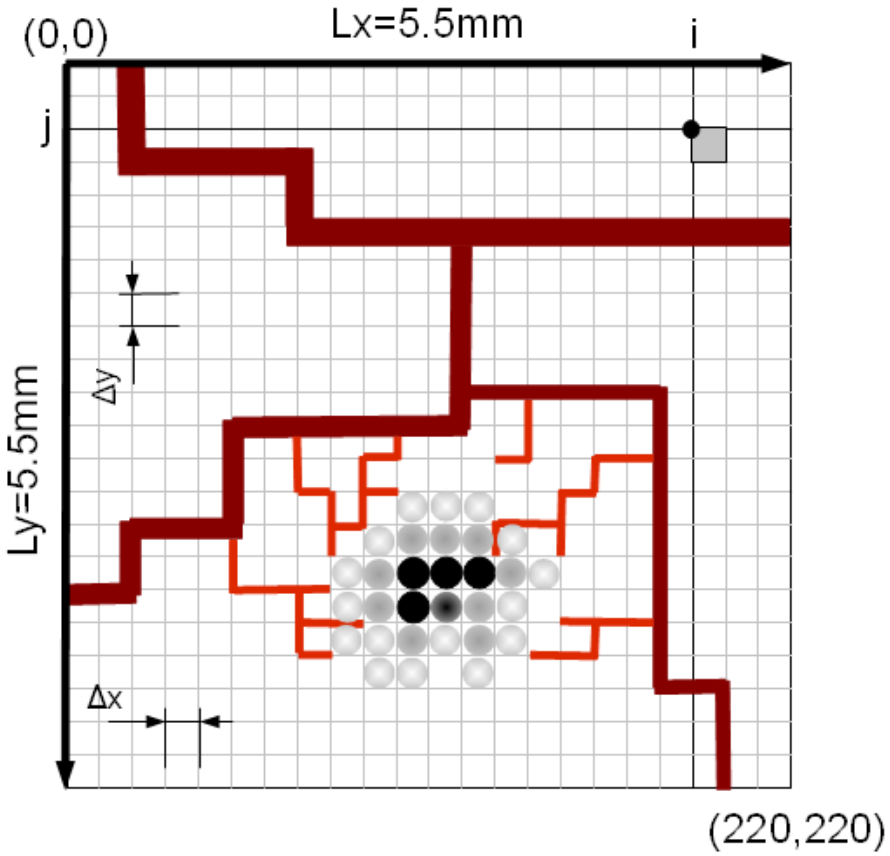
$$\frac{\partial O}{\partial t} = D_O \nabla^2 O + \gamma_v W_{i,j} (O_v - O) - k_{i,j} O$$

Vessels "weight" *kn, kp, kq*

The computational model

Tumour growth, metastatic invasion
Cell cycle, metabolism and death

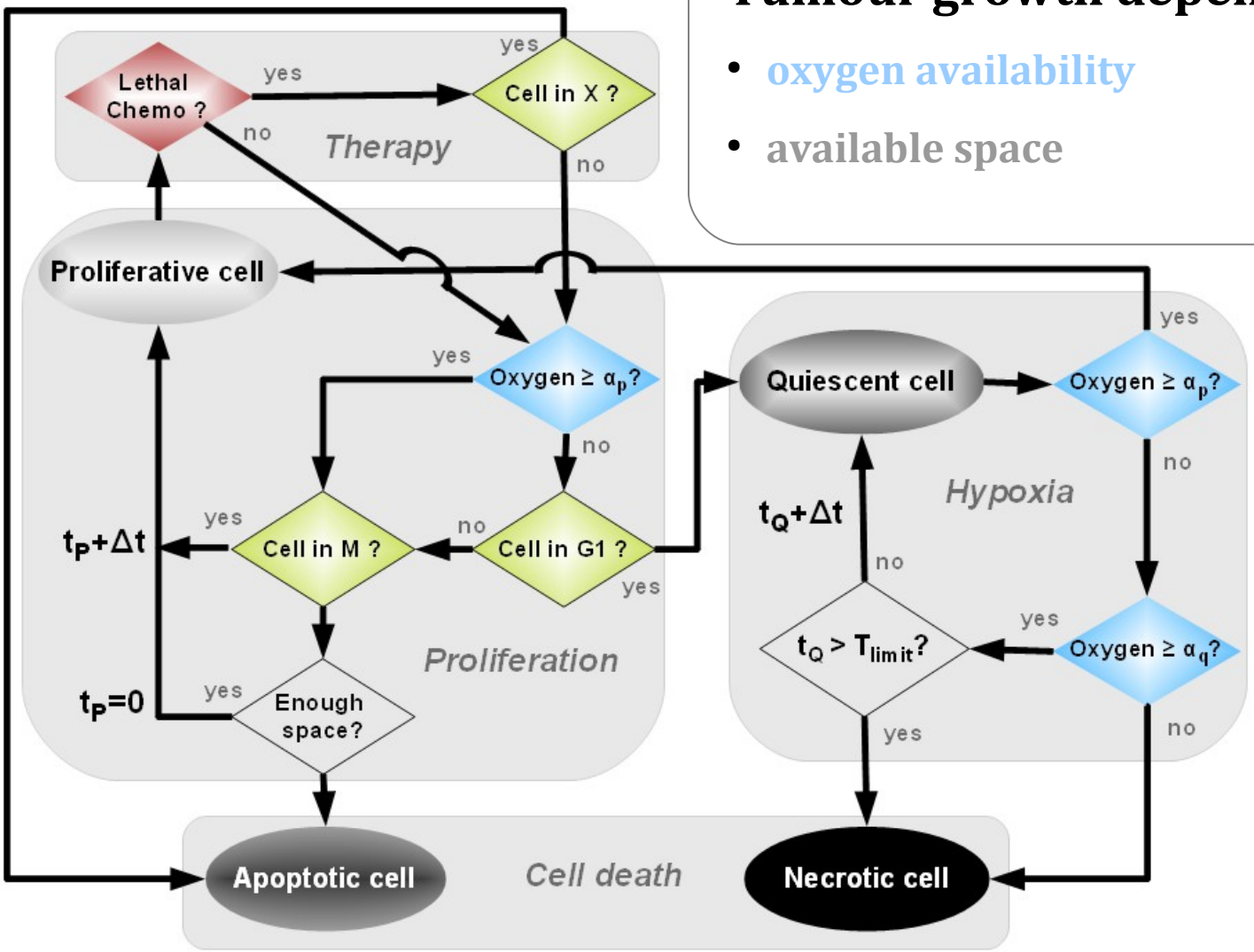
Angiogenesis, vascular adaptation
Vessels permeability, diffusion of therapeutic molecules and oxygen

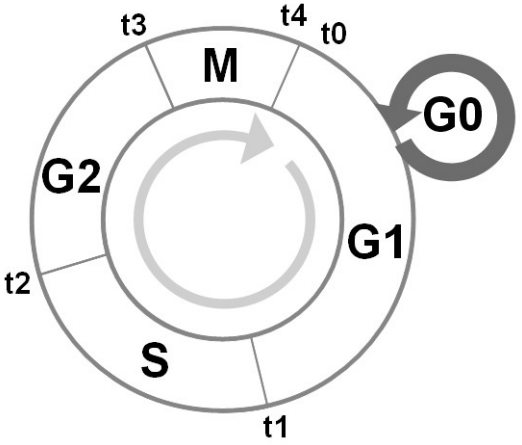


Modelling tumour growth

Tumour growth depends on:

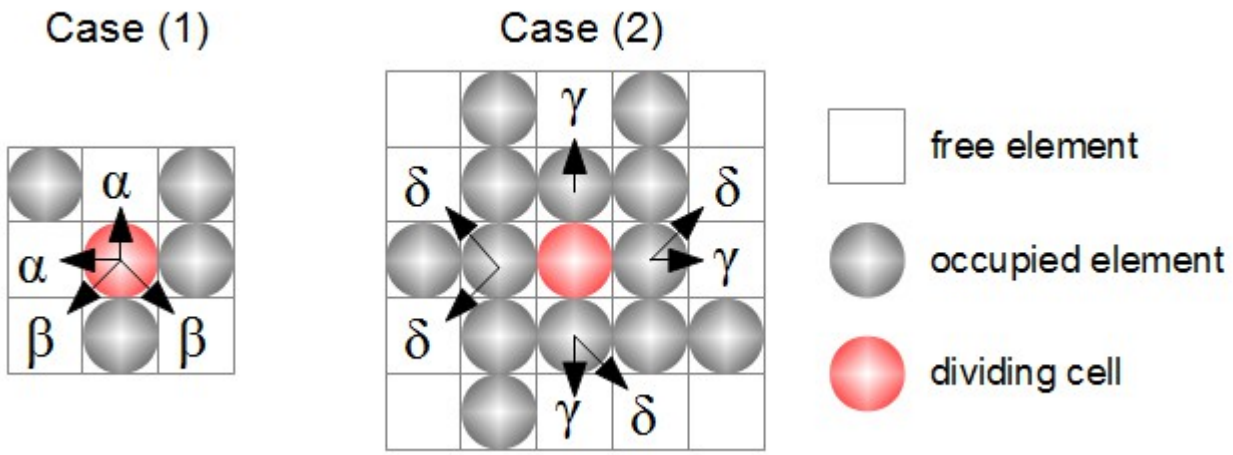
- oxygen availability
- available space
- cell cycle
- concentration of cytotoxic molecules





Modelling tumour growth

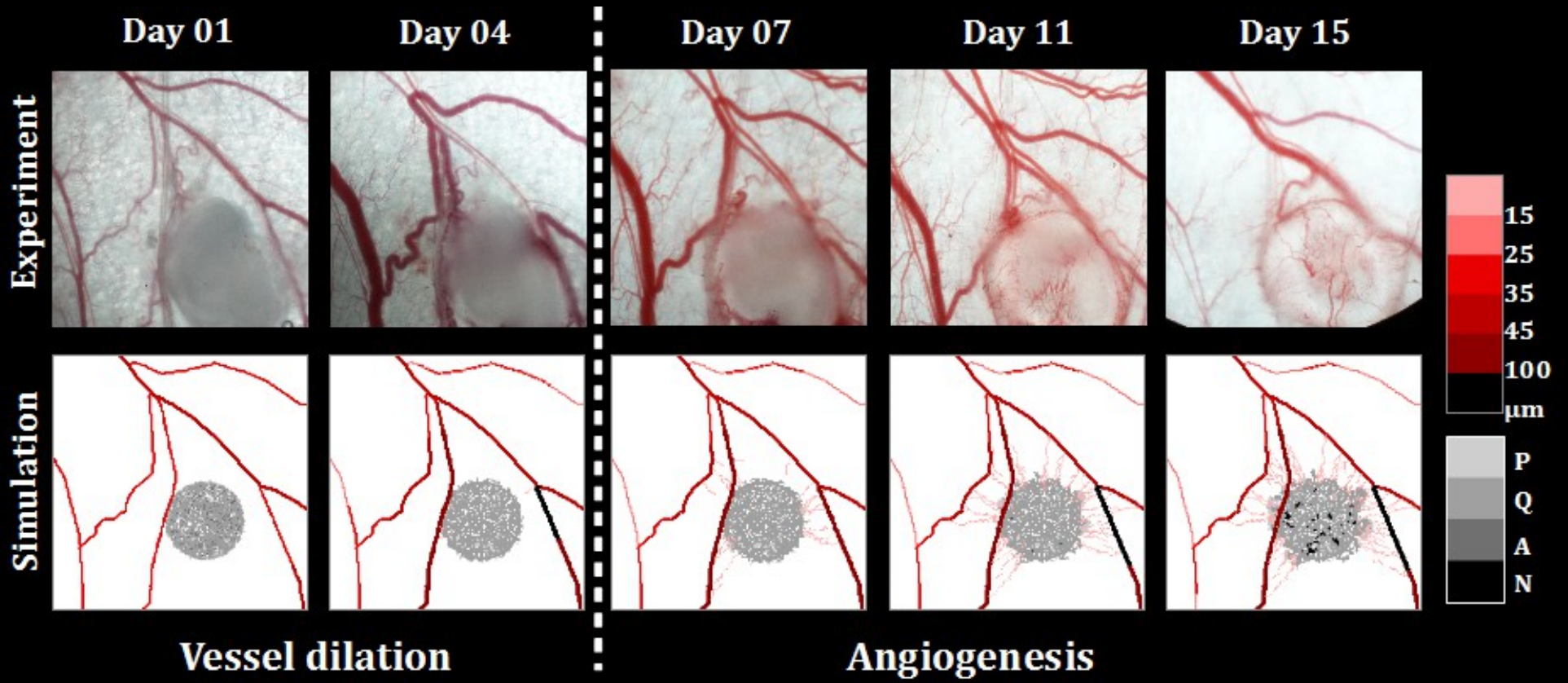
Rules for cell division



$\beta < \alpha$ $\delta < \gamma$

Model validation with experimental model (1)

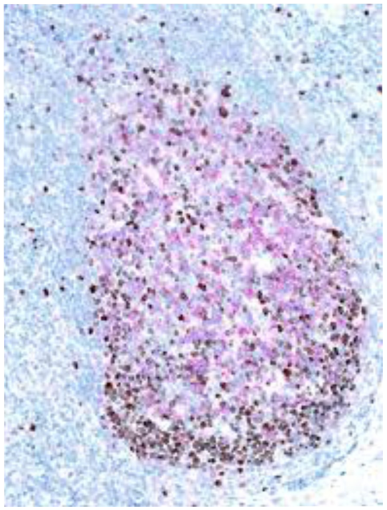
- ✔ **Comparison of simulated and observed kinetics of tumour development**
- ✔ Comparison of virtual and histological tumour slices
- ✔ Adjustment of parameters and validation of hypotheses



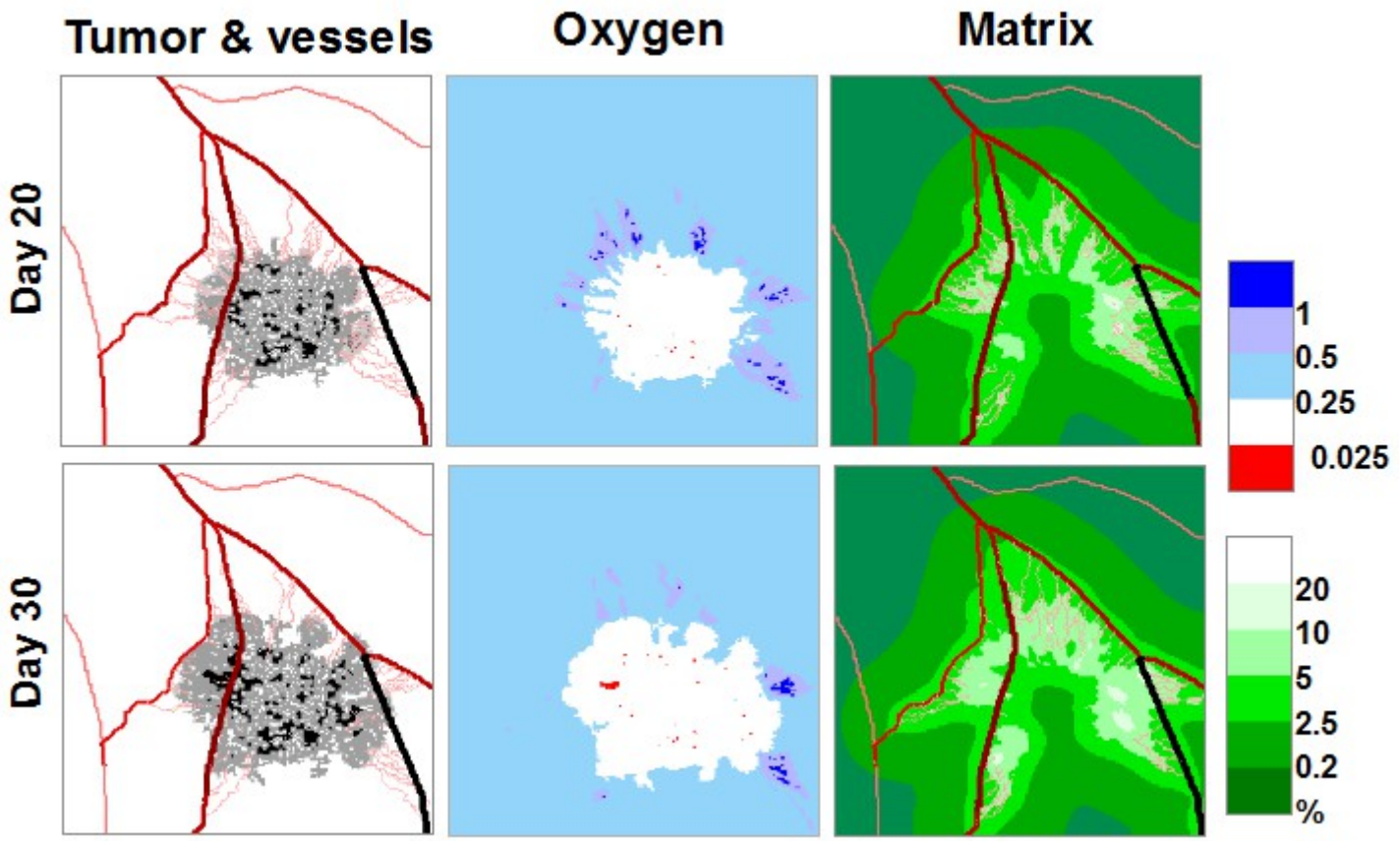
Anne-Cécile Lesart, PhD Thesis, 2013

Model validation with experimental model (1)

- ✔ Comparison of simulated and observed kinetics of tumour development
- ✔ **Comparison of virtual and histological tumour slices**
- ✔ Adjustment of parameters and validation of hypotheses



Histological slice



Anne-Cécile Lesart, PhD Thesis, 2013

Model validation with experimental model (1)

✔ Comparison of simulated and observed kinetics of tumour development

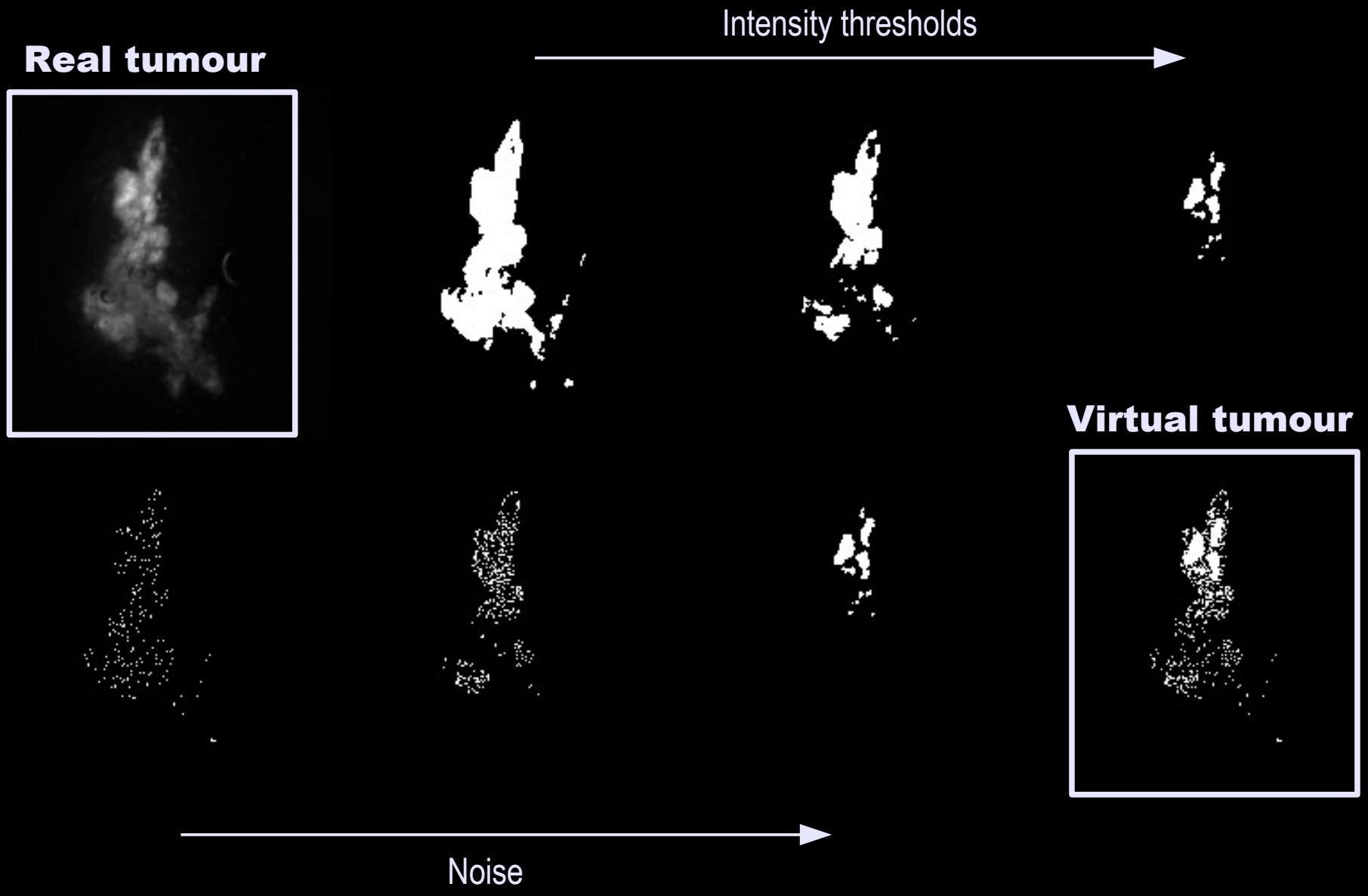
✔ Comparison of virtual and histological tumour slices

✔ **Adjustment of parameters and validation of hypotheses**

Parameters adjusted

- Durations of cell cycle phases and level of variability
- Oxygen thresholds for transition to quiescence (hypoxia) and for cell death
- Oxygen consumption rates for each cell type
- Vessels permeability to oxygen
- Diffusion, production/consumption rates of vascular proteases

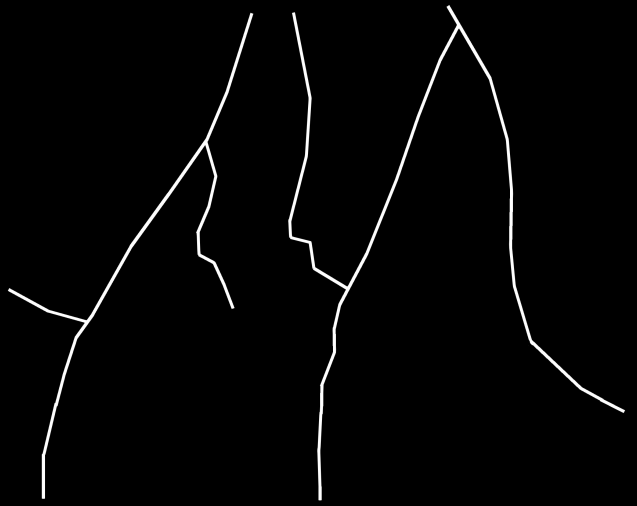
Initialization of the tumour with experimental model (2)



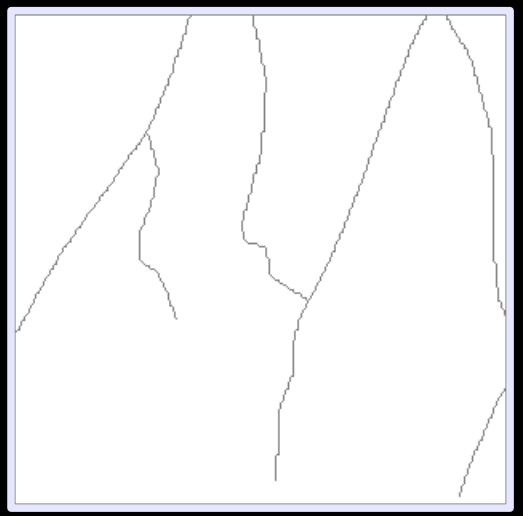
Initialization of the vasculature with experimental model (2)

Bright field image

Vascular skeleton

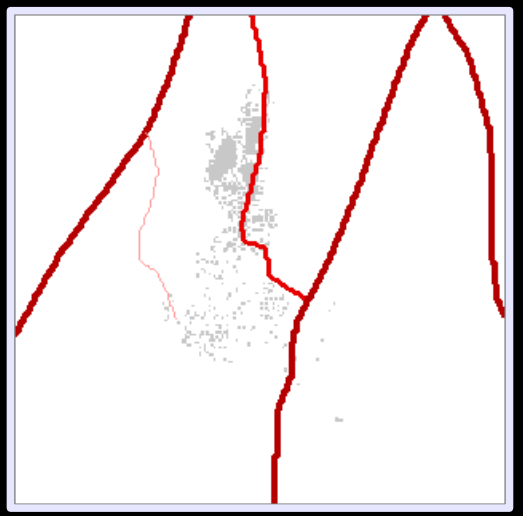


Cropped image

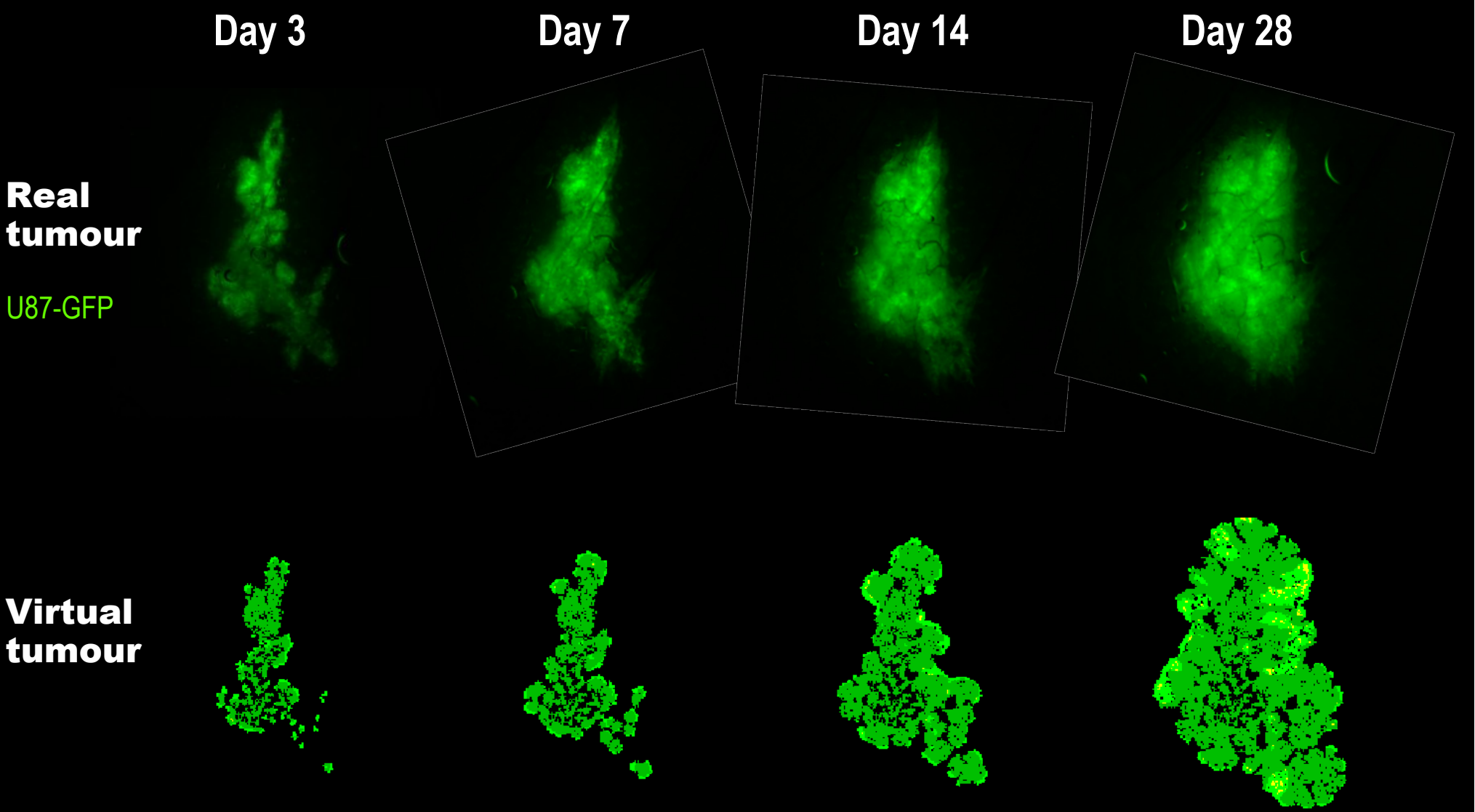


Perfusion

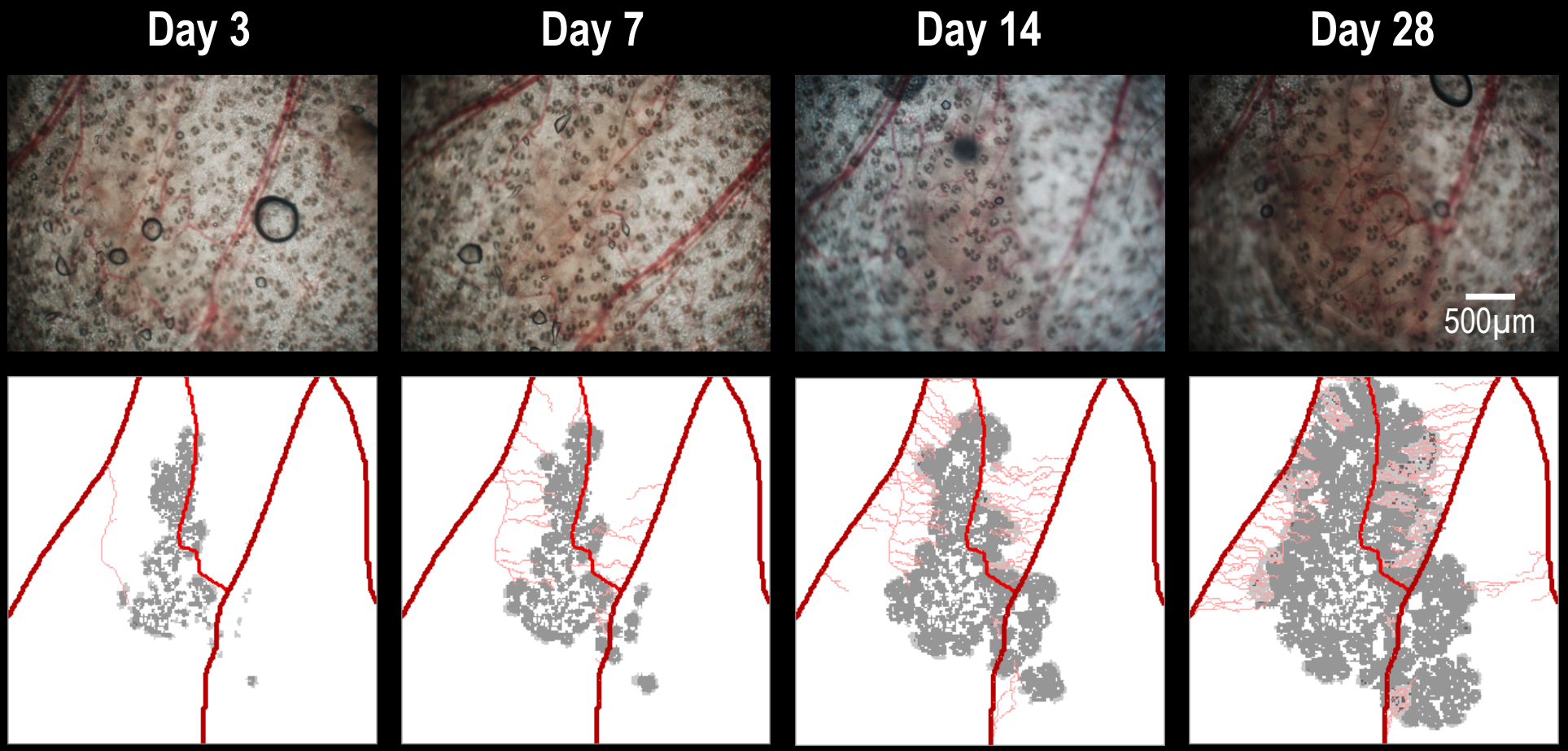
Initial vasculature + tumour



Real *vs* virtual tumour

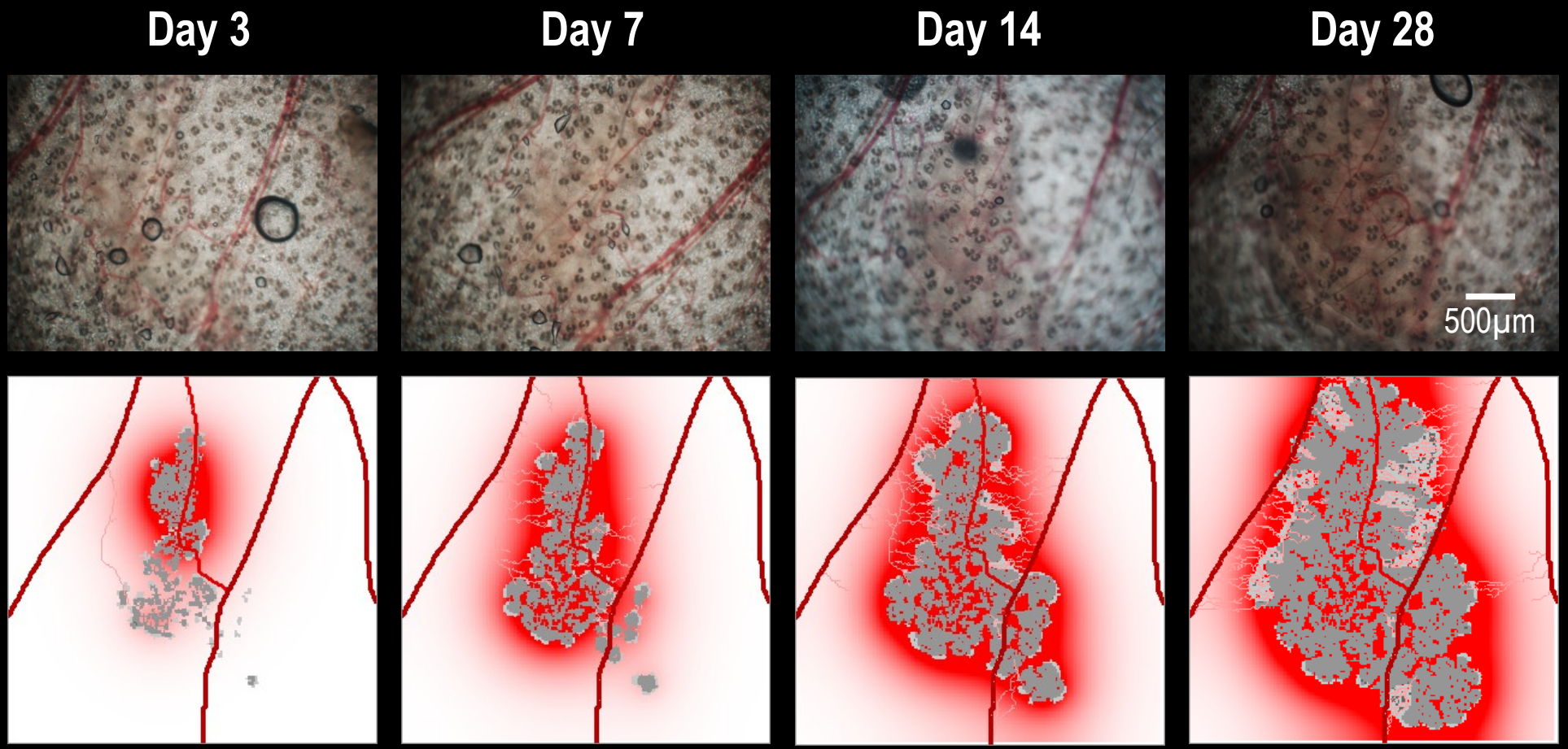


Real *vs* virtual tumour



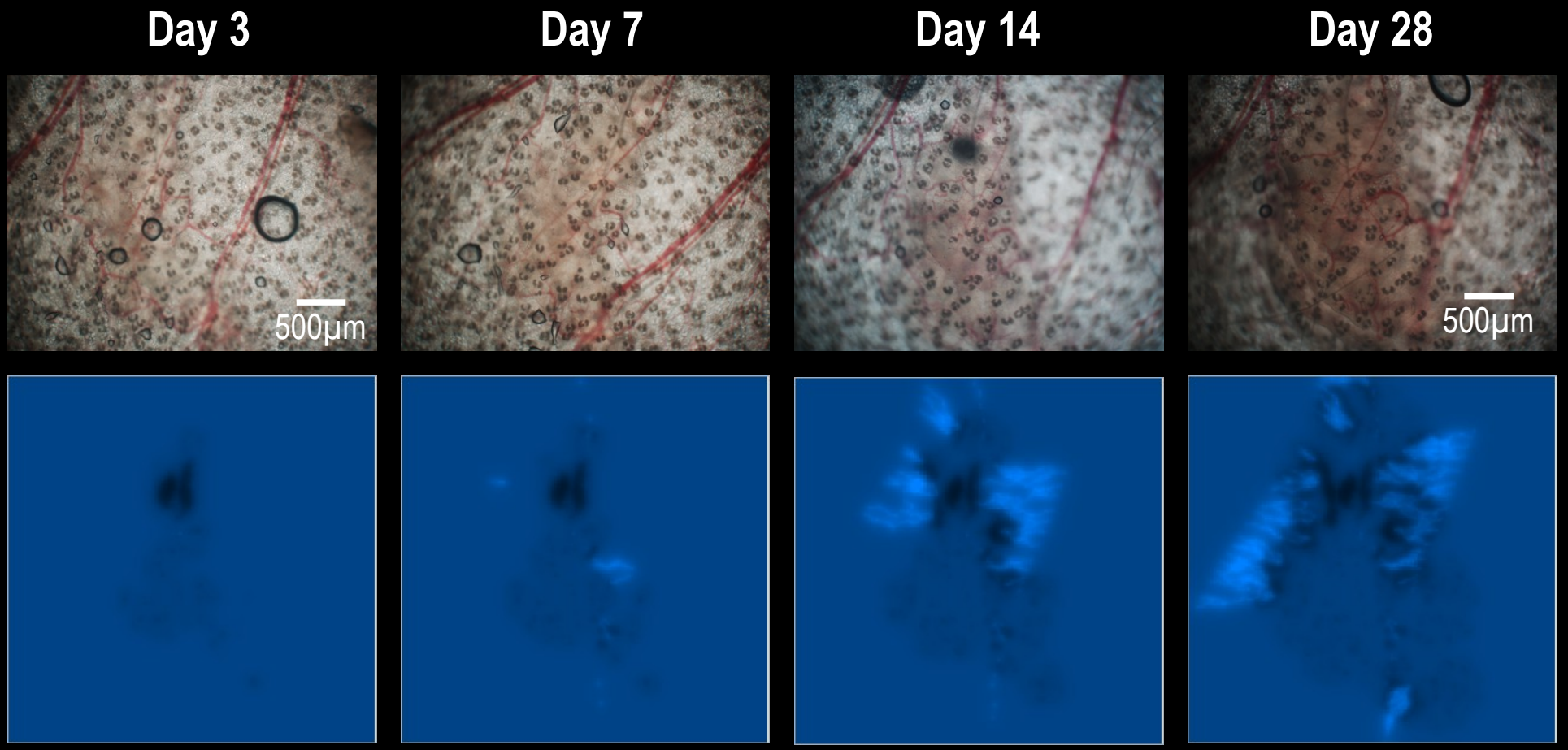
Angiogenesis

Real *vs* virtual tumour



Angiogenesis + VEGF diffusion

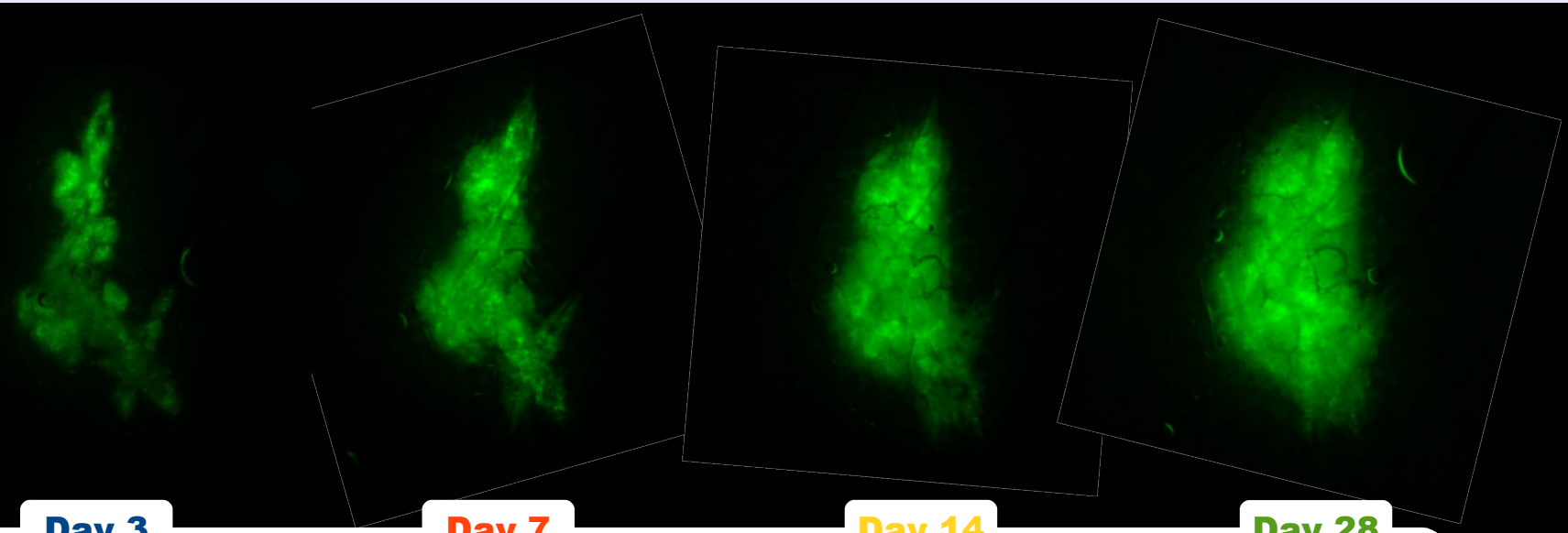
Real *vs* virtual tumour



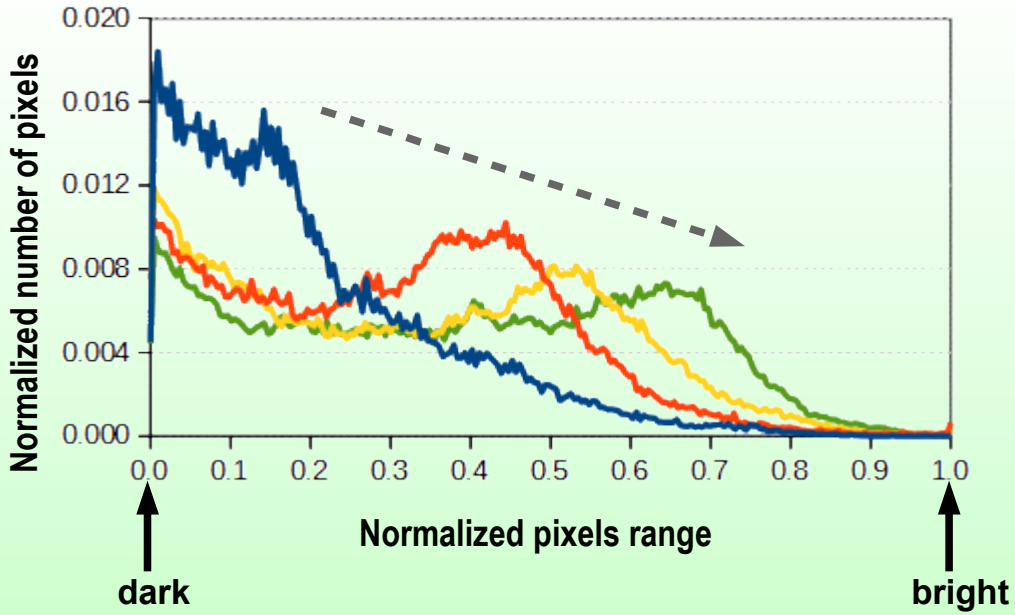
Tumour growth : texture and size

Real tumour

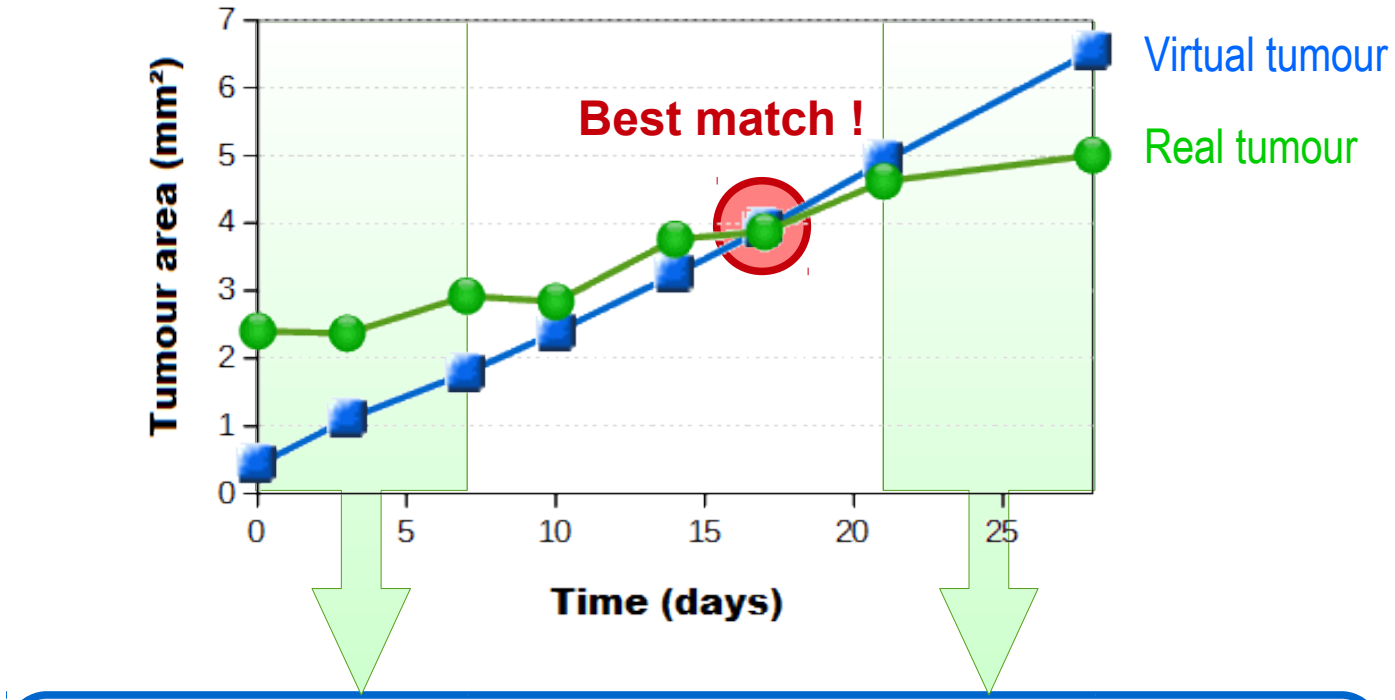
U87-GFP



Day 3 Day 7 Day 14 Day 28



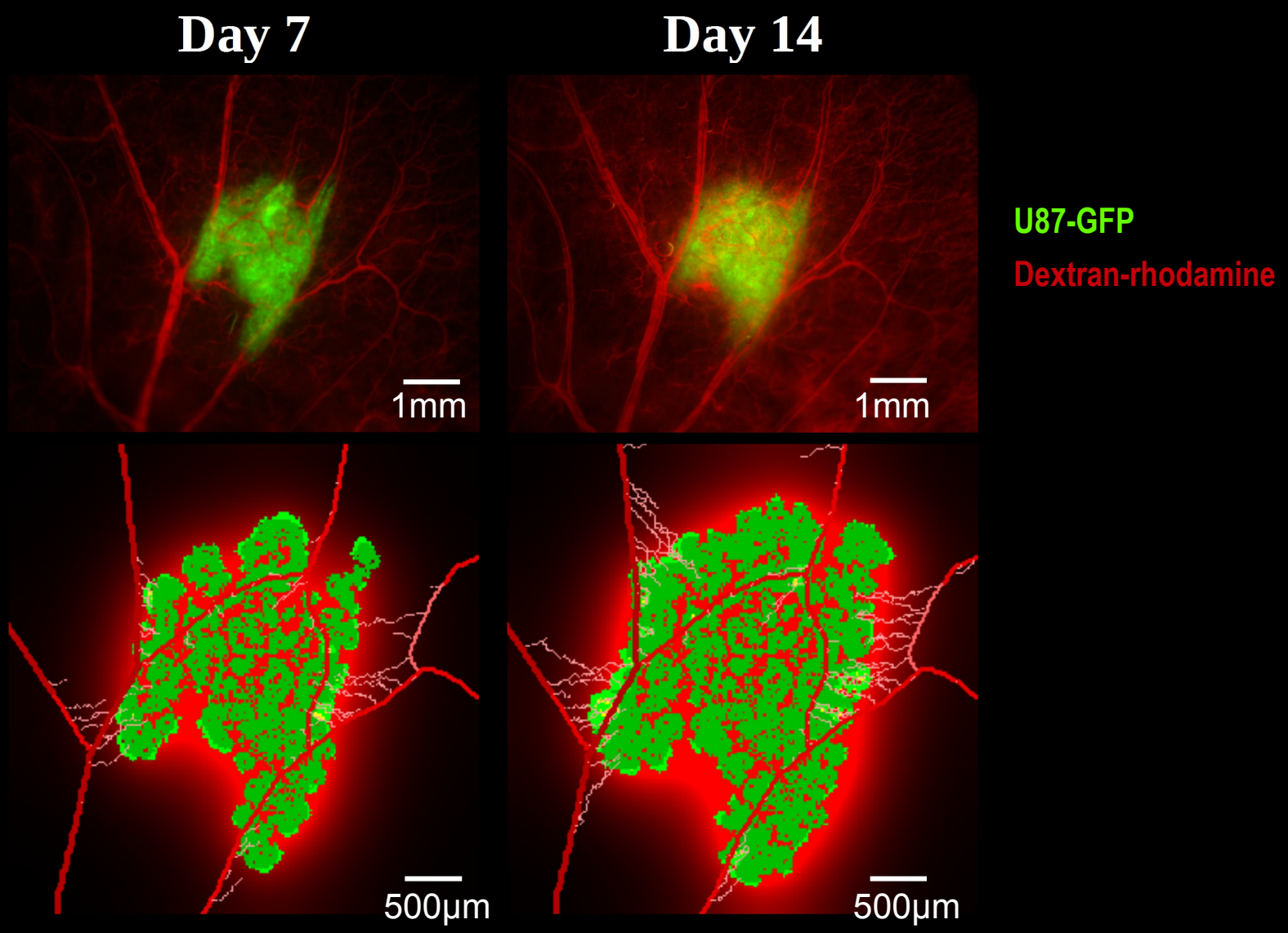
Tumour growth : texture and size



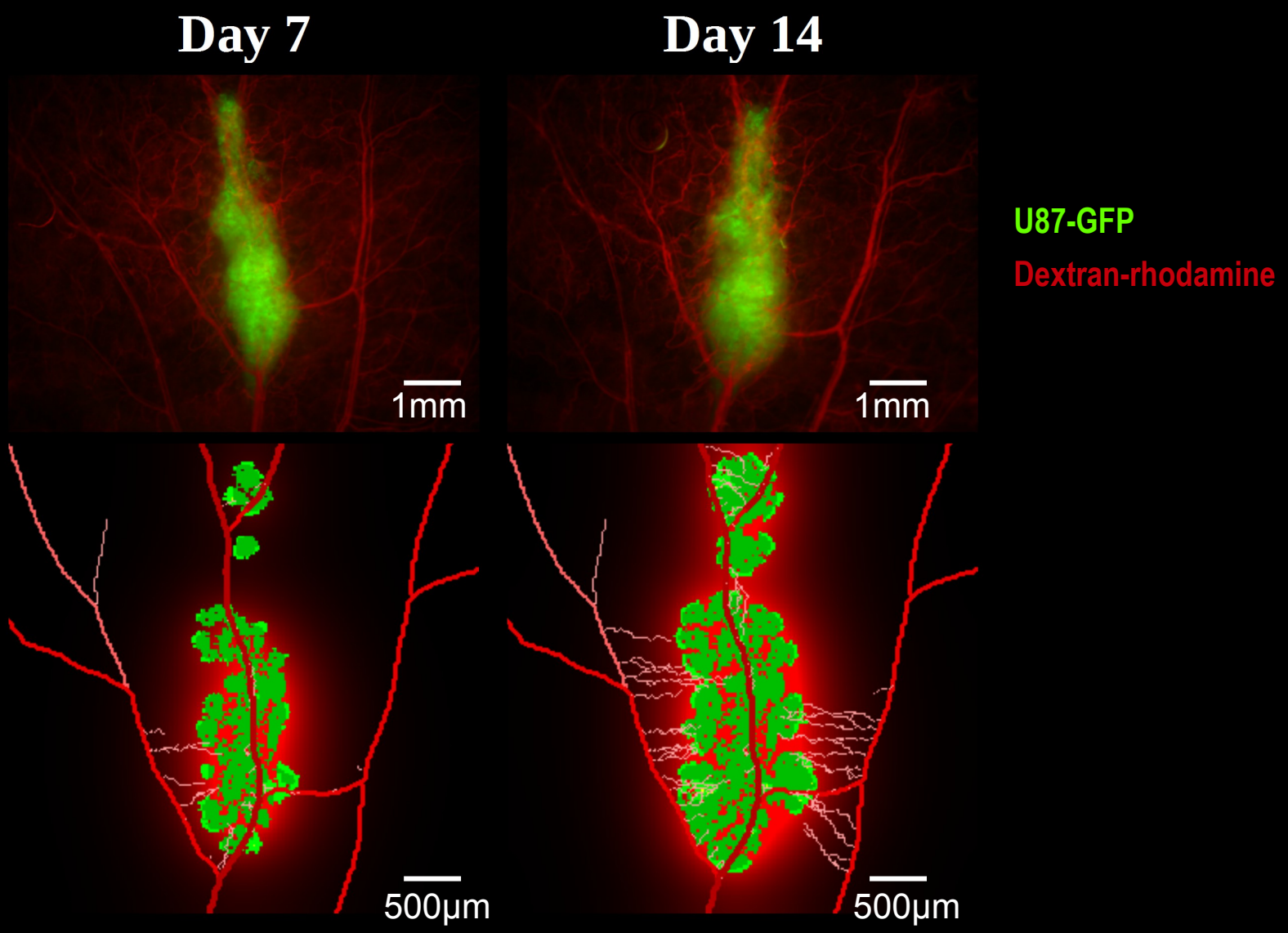
2D slice (virtual)
vs
3D integration (real)

2D spreading (virtual)
vs
3D compaction (real)

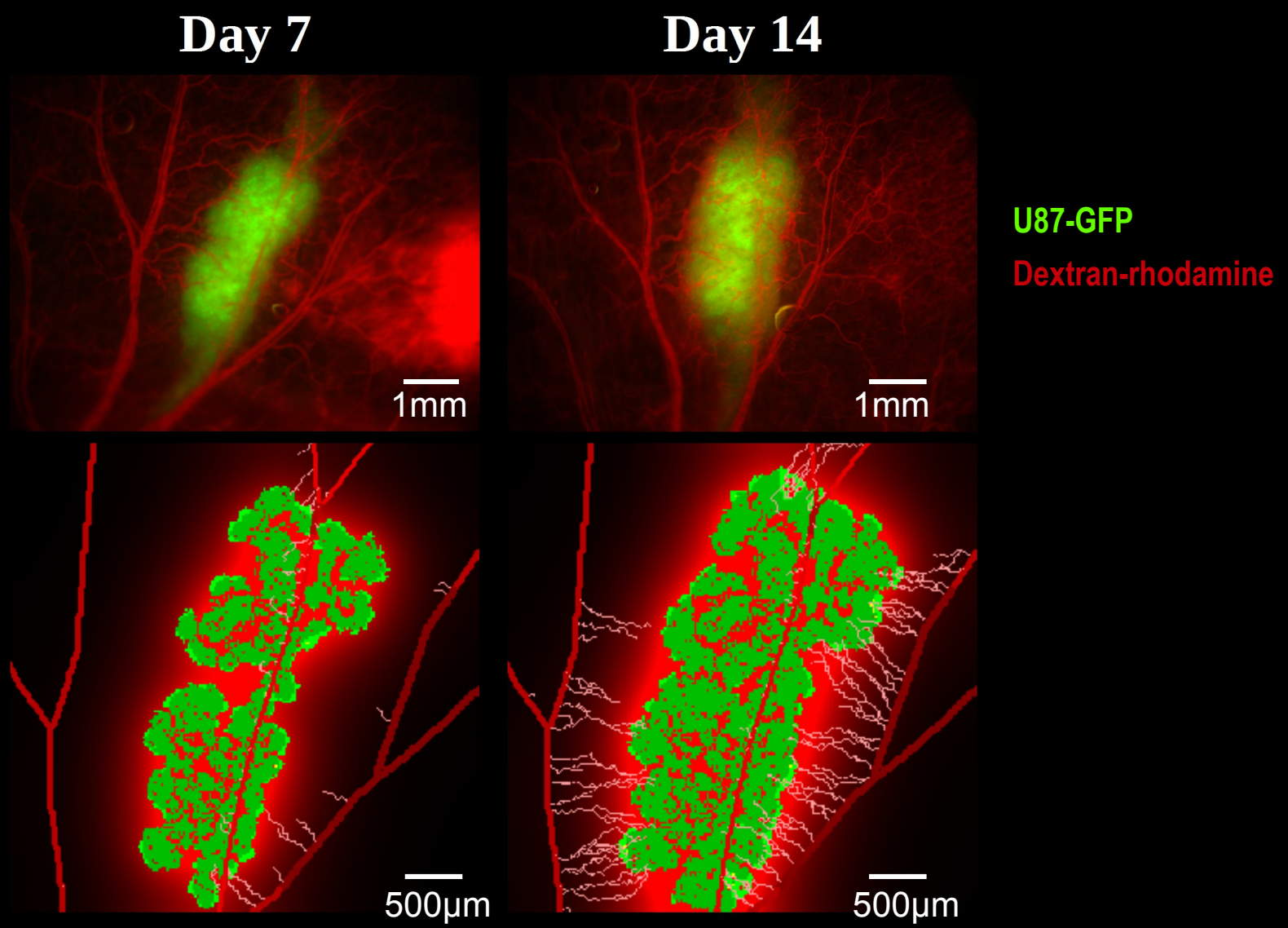
Tumour development in one week (case 1)



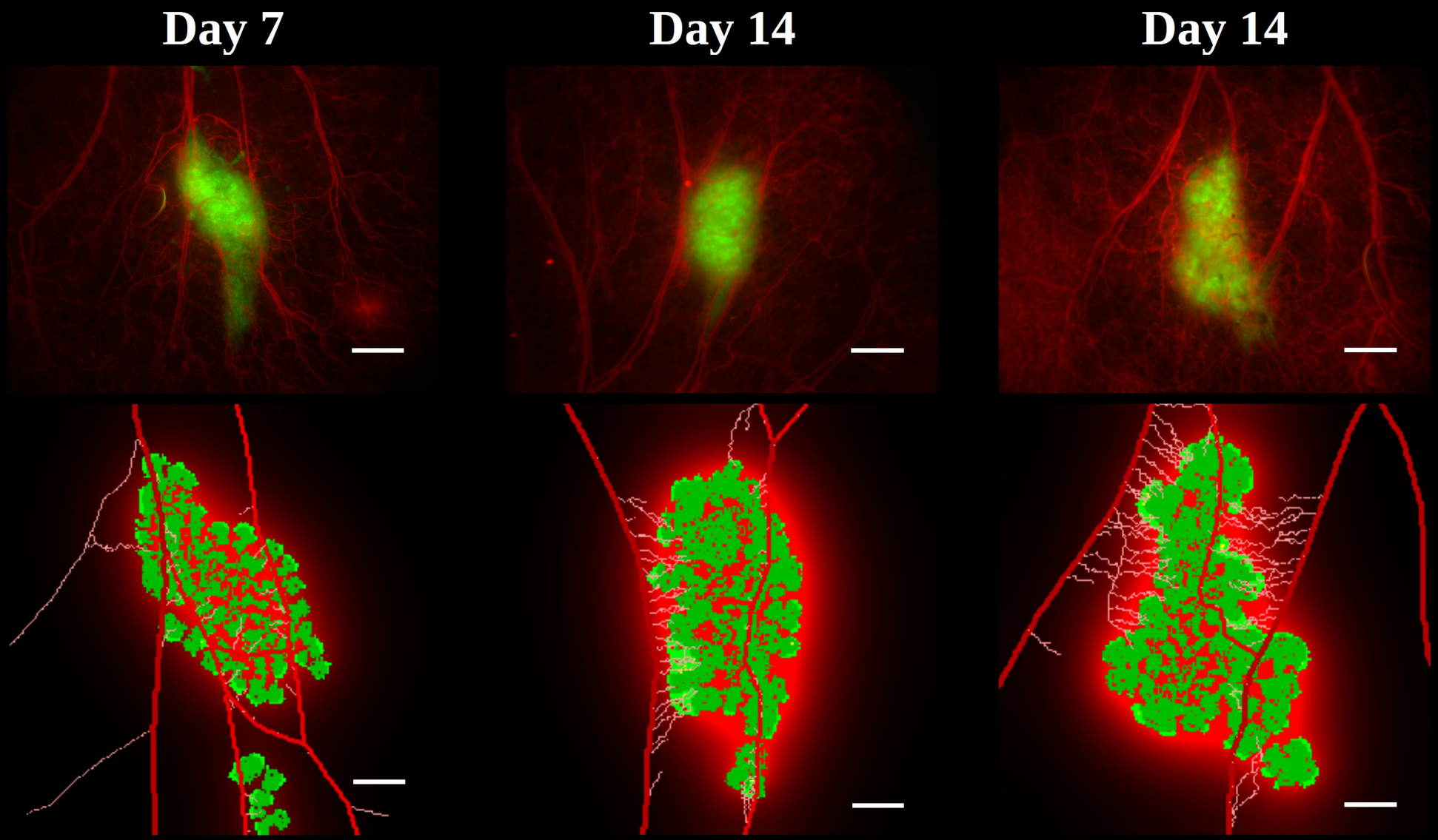
Tumour development in one week (case 2)



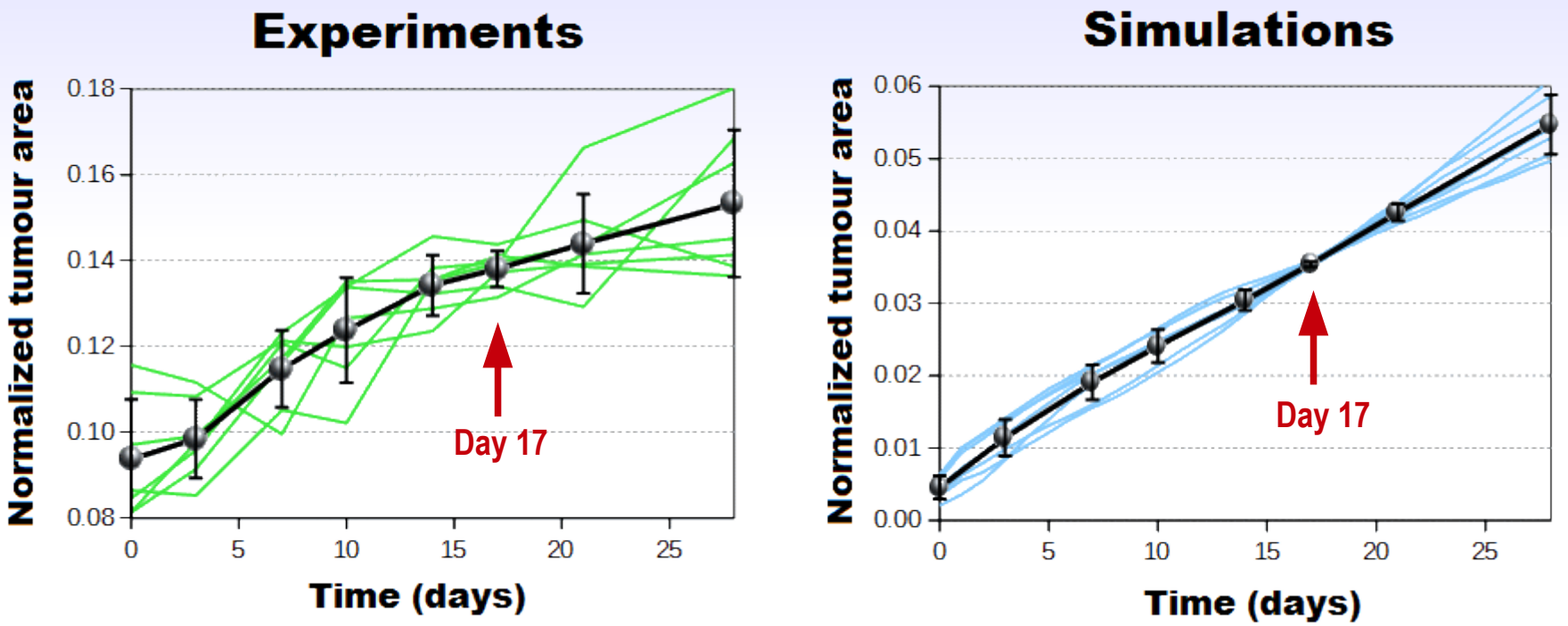
Tumour development in one week (case 3)



Tumour development (other cases)



The *angiogenic bottleneck*



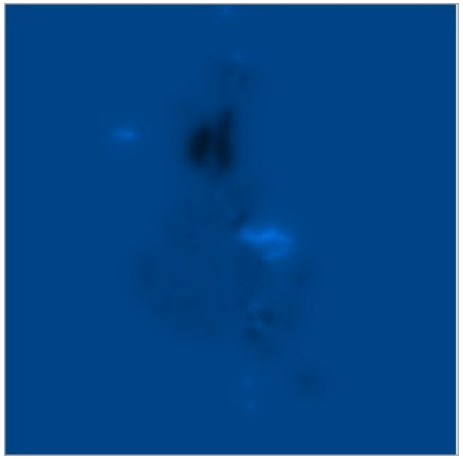
Slower growing tumours have a *higher angiogenic potential* since they possess a higher proportion of hypoxic cells and reciprocally.

Convergence of the growth curves on Day 17

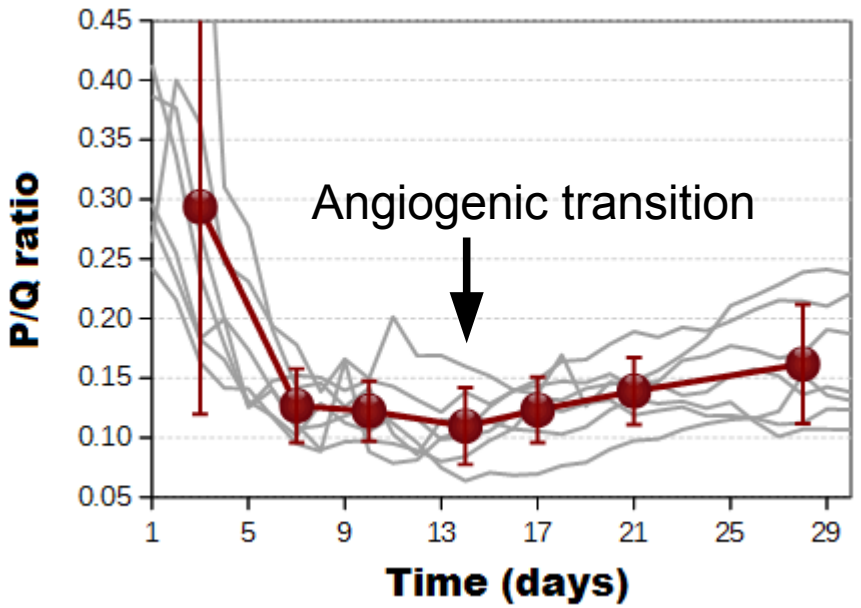
The *angiogenic bottleneck*

Proliferative *vs* quiescent cells

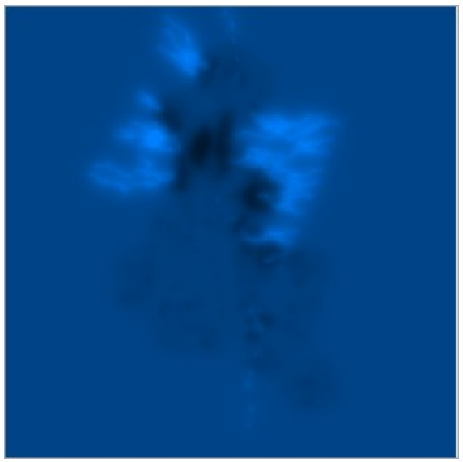
Day 7



Low oxygen level



Day 14

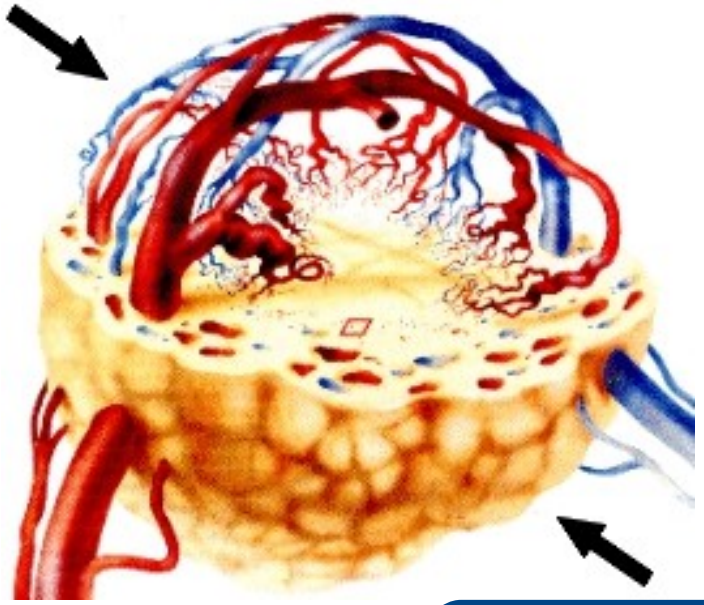


high oxygen level

Therapies

Vascular disrupting agents

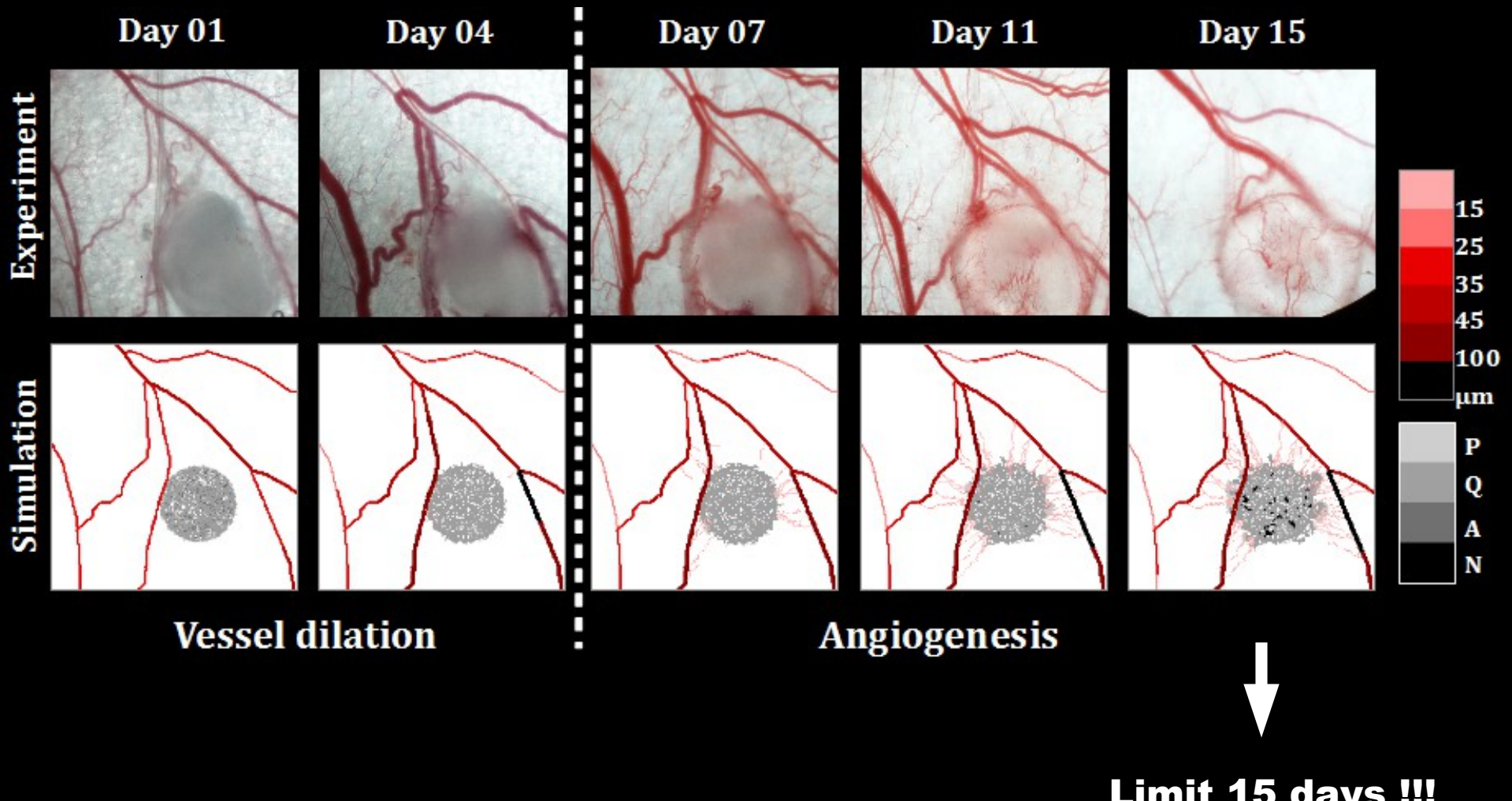
target = vessels



Cytotoxic molecules

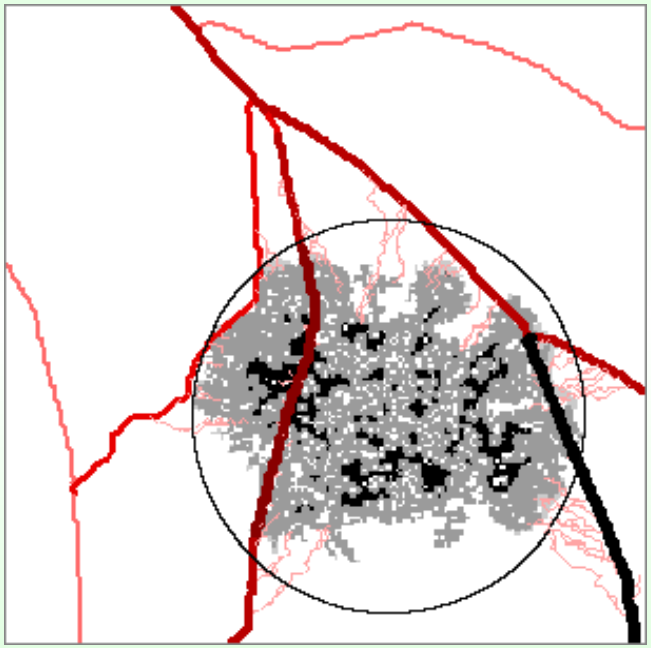
target = tumour cells

Simulation of growth without treatment



Simulation of cytotoxic effects

Without treatment



30 days

The protocol is defined by :

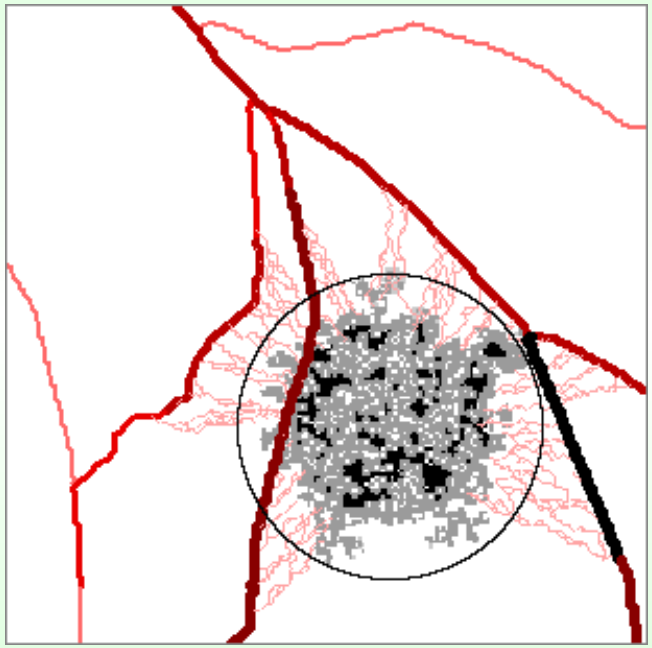
- The choice of the molecule (target phase)
- The dose administered
- The duration of administration
- The frequency of administration

Anne-Cécile Lesart, PhD Thesis, 2013

Simulation of cytotoxic effects

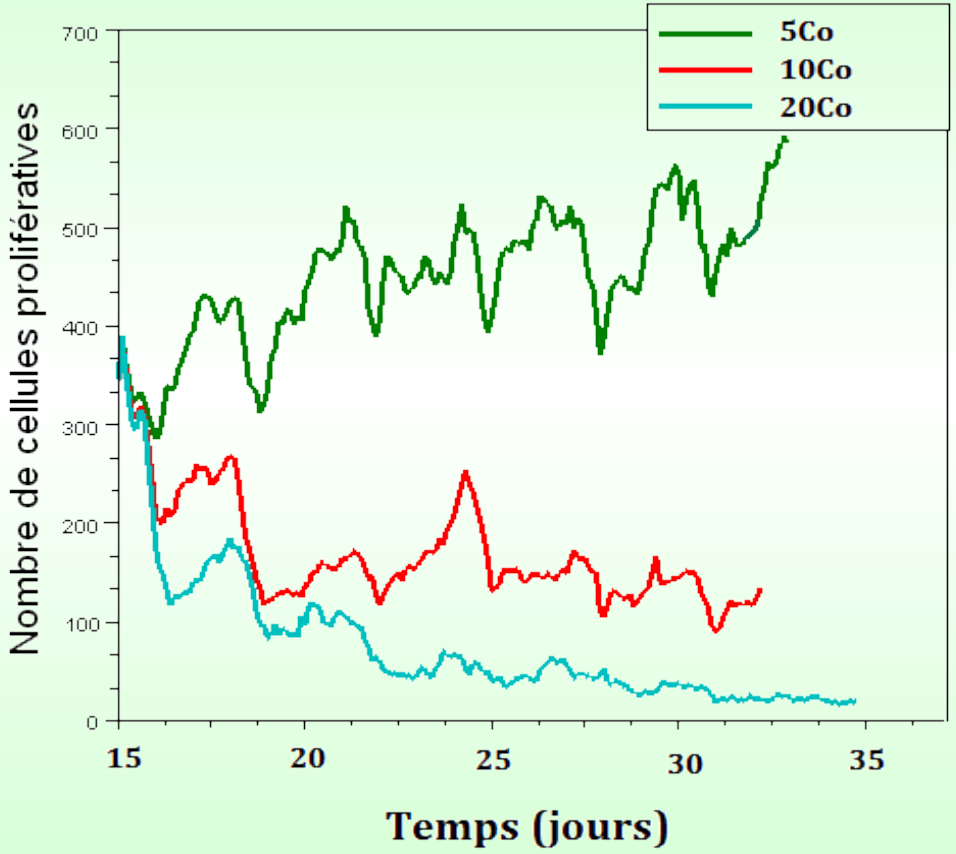
Dose variation

With cytotoxics



30 days
(after 15 days of treatment)

duration = 2h, frequency = 3 days

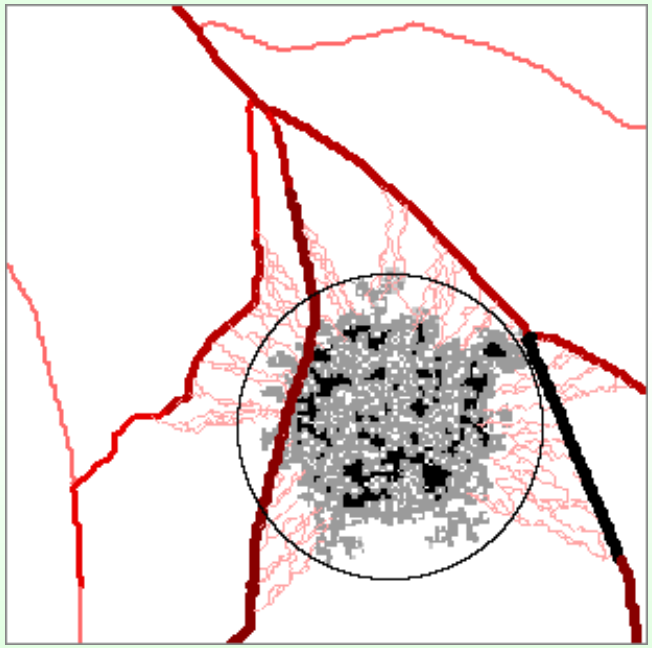


Anne-Cécile Lesart, PhD Thesis, 2013

Simulation of cytotoxic effects

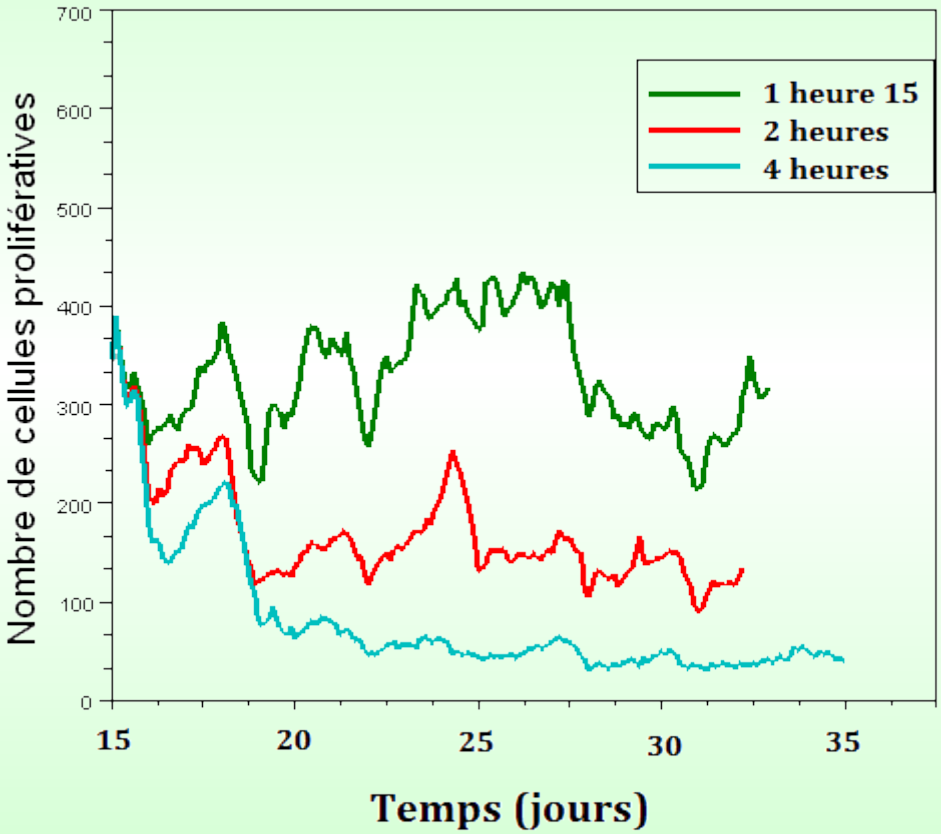
Variation of the duration

With cytotoxics



30 days
(after 15 days of treatment)

dose = $10C_0$, frequency = 3 days

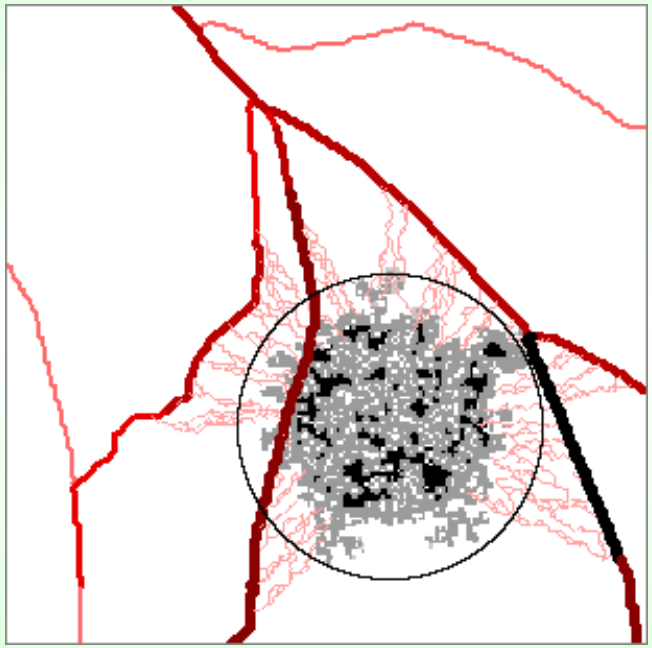


Anne-Cécile Lesart, PhD Thesis, 2013

Simulation of cytotoxic effects

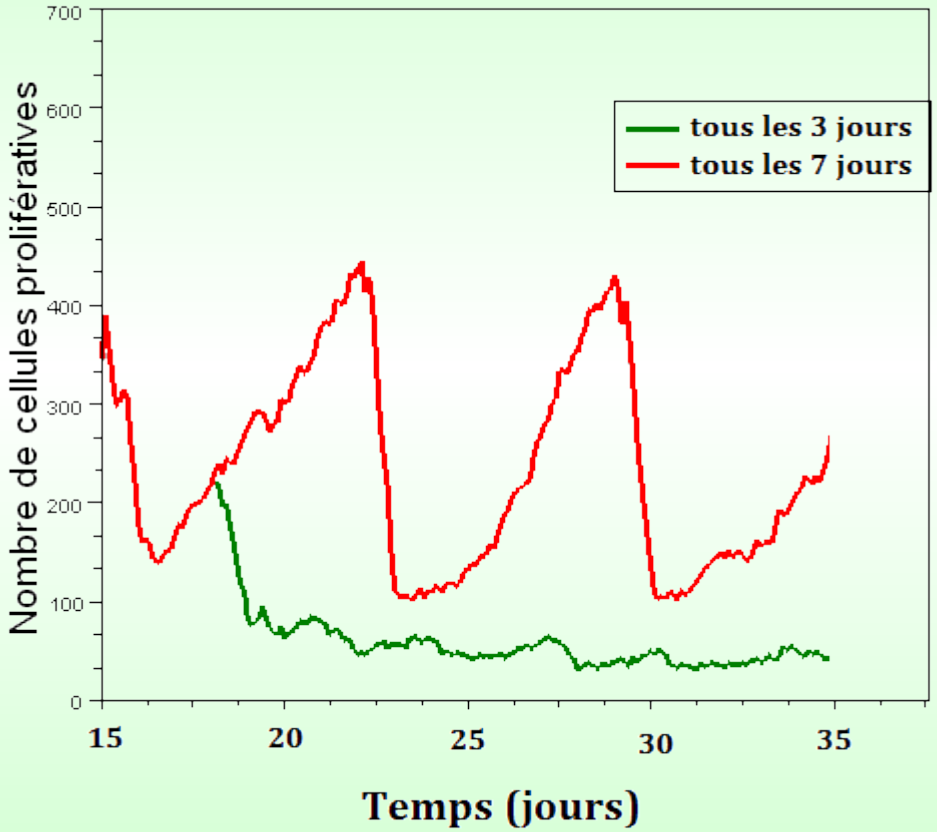
Variation of the frequency

With cytotoxics



30 days
(after 15 days of treatment)

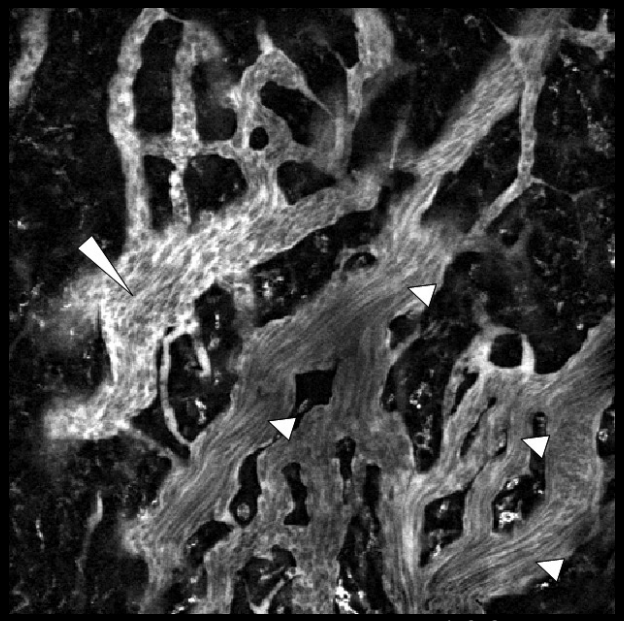
dose = $10C_0$, duration = 4h



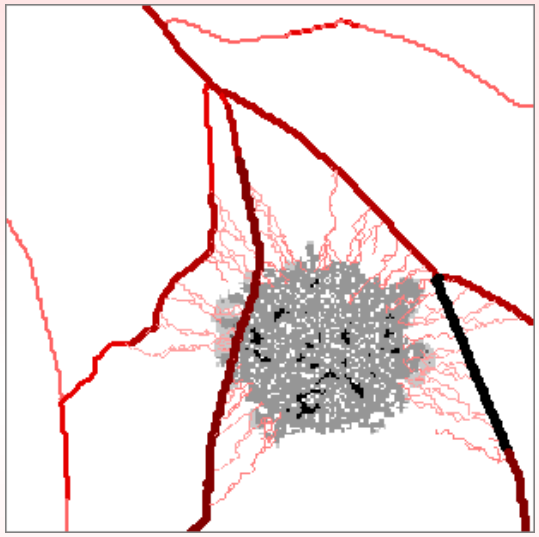
Anne-Cécile Lesart, PhD Thesis, 2013

Simulation of Vascular Disrupting Agents (VDA)

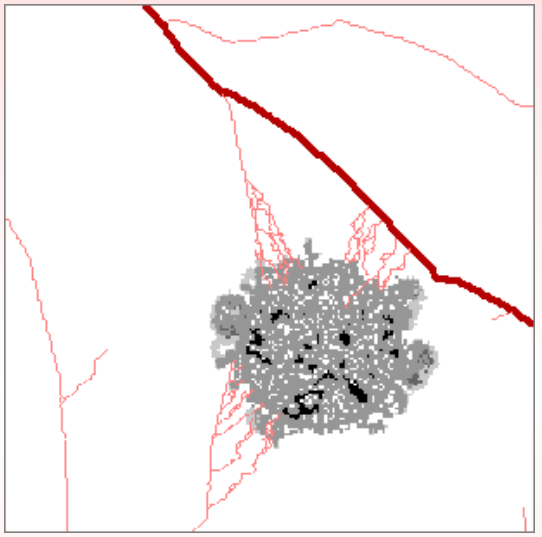
Brutal arrest of blood flow a few seconds after the administration of Vascular Disrupting Agents



Before VDA



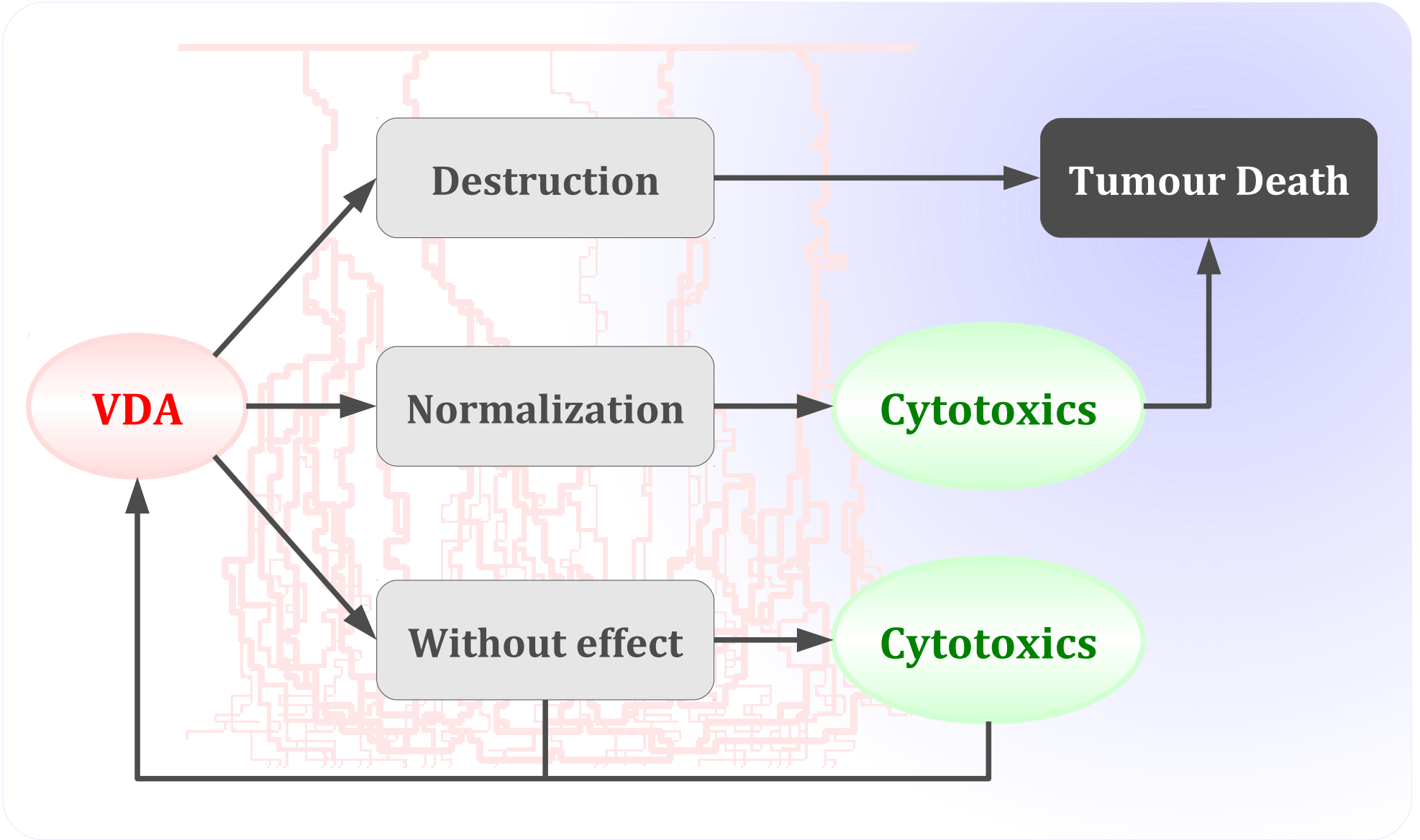
After VDA



- massive necrosis
- resistant or more invasive phenotypes

Anne-Cécile Lesart, PhD Thesis, 2013

Therapeutics coupling



The next step ...

	Phase 1	Phase 2
Experimental model	Sub-cutaneous implantation of U87-GFP tumours cells in matrigel *Nude mice (immuno-deficient)*	Intra-cerebral implantation of C6 or 9L tumour cells *Wistar rat*
		
Virtual model	2D isotropic tissue	3D anisotropic brain tissue
Control	Intravital two-photon microscopy, immuno-histology	MRI, immuno-histology
Action	Cytotoxics and VDA	Cytotoxics, VDA and radiotherapy

Partners

Techniques for biomedical engineering and complexity

Cell & tissue dynamics and functional microscopy

Nicolas Glade, Arnaud Chauvière (computational modelling)

Marie-Paule Montmasson, Malika Hamel (experimental cell models, histology)

Arnold Fertin, Yves Usson (image analysis)

Clinatec *Biomedical research centre*

Flavien Caraguel , Boudewijn van der Sanden (*in vivo* models, intavital imaging)

Grenoble Institute of Neurosciences

Emmanuel Barbier, Benjamin Lemasson (brain tumour models, MRI)

François Estève (brain tumours, radiotherapy)

Grenoble Images, Speech Signal and Control

Mazen Alamir, Mirko Fiacchini (control theory and optimization)

Ecrins Therapeutics

Andrei Popov, Aurélie Juhem (development of antivascular molecules)

Abstract

The design of a patient-specific virtual tumour is an important step towards personalized medicine since the virtual tumour can be used to define the most adapted and efficient treatment protocol. However this requires to capture the description of many key events of tumour development, including angiogenesis, matrix remodelling, hypoxia, cell heterogeneity that will all influence the tumour growth kinetics and degree of tumour invasiveness. To that end, an integrated hybrid and multiscale approach has been developed based on data acquired on a preclinical mouse model as a proof of concept. Fluorescence imaging is exploited to build case-specific virtual tumours and to validate their spatiotemporal evolution. The validity of the model will be discussed as well as its potential to identify the best therapeutic strategy for each individual tumour case.