nanoHUB Account

- This talk's online PhysiCell models are cloud-hosted on nanoHUB.org.
- nanoHUB is **free**, but it requires a one-time registration.
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  3. Choose a Google account
  4. Click "No" (so it doesn't try to associate with some other nanoHIB account)
  5. Finish filling in details, and you're done!
  6. Use your google account to sign in in the future.

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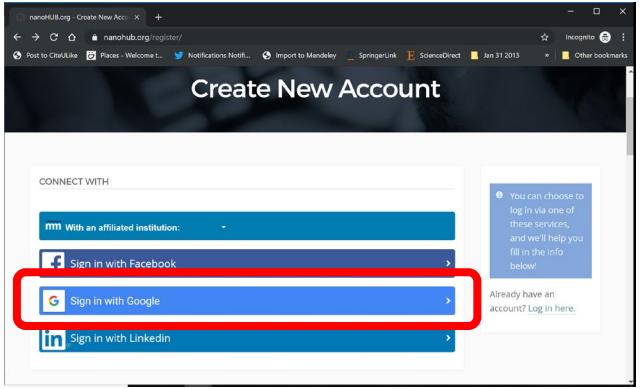

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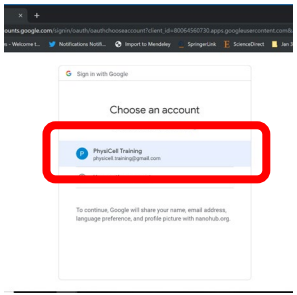

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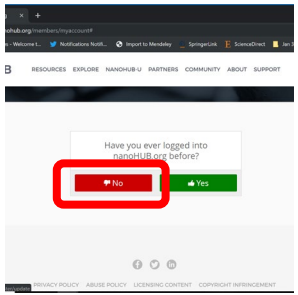

How Simple Cell-Cell Interactions Lead to Complex Multicellular Dynamics

Paul Macklin, Ph.D.

Intelligent Systems Engineering
Indiana University

December 20, 2020

Thanks: Partners

- **Colon cancer metabolic cross-talk in organoids:**
  - Stacey **Finley** (USC, U01 Contact PI)
  - Shannon **Mumenthaler** (USC, U01 PI)
- **Hypoxia in breast cancer invasion:**
  - Daniele **Gilkes** (JHU, JKTGF Contact PI)
- **ECM and leader-follower interactions:**
  - Andy **Ewald** (JHU, BCRF & JKTGF Contact PI)
  - Newton (USC), Peyton (Umass), Bader (JHU)
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  - **alumni:** S.H. Friedman, A. Ghaffarizadeh (USC)
- **High-throughput on HPC:**
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  - Chase Cockrell, Gary **An** (University of Vermont)
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  - Boolean network extensions: Institut Curie (Letort, Montagud, Stoll, Barillot, Zinovyev, Calzone)
  - Flux balance extensions: Miguel Ponce de Leon (Barcelona Supercomputing Center)
  - GPU computing prototypes: Sunita Chadrasakaran (Delaware)
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- **IU PhD students:**
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  - Yafei Wang (liver mets, nanotherapy)
  - Furkan Kurtoglu (multicellular metabolism)
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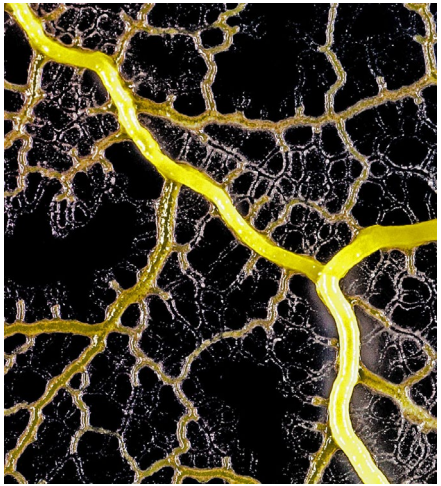
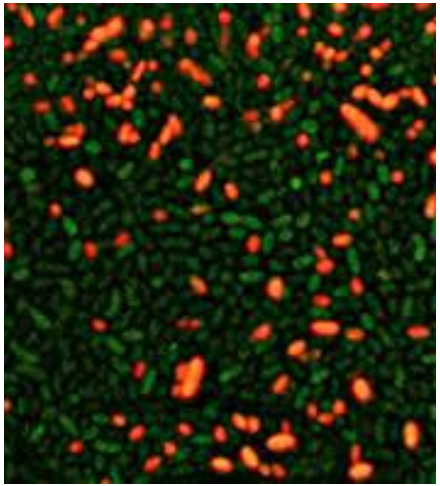
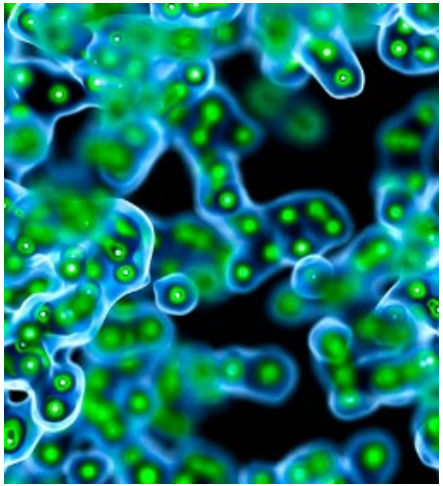
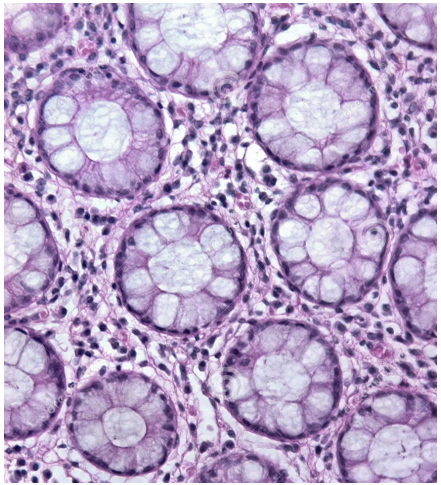
Simple single-cell behaviors …

- **growth**: Cell volume increases as it progresses through the cell cycle.
- **division**: Cells reproduce by dividing in half into daughter cells.
  - These cells may or may not be identical!
- **death**: Cells can die by a variety of mechanisms. They don't disappear immediately, but instead shrink and continue to interact with other cells.
  - **apoptosis:** An orderly, planned shutdown process (that still requires and consumes energy!)
  - **necrosis:** A disorderly, unplanned shutdown process, often from energy depletion.
- **adhesion**: Cells can stick to other cells or to fibers in their environment.
- **resistance to deformation**: Cells are largely incompressible and viscoelastic. They exert resistant forces on other cells and the environment.
- **motility**: Cells actively move through the environment in a biased random walk.
- **secretion**: Cells can secrete chemical factors to communicate.
- **uptake**: Cells can consume chemical factors (e.g., oxygen)
- **sampling**: Cells can sample chemical factors and other physical properties in their immediate surroundings.

Give rise to complex systems

- ***Multicellular systems***—composed of multiple cells of multiple types—can exhibit remarkable diversity, with complex emergent behaviors.

How do these systems self-organize and sustain themselves?

How do we understand these multiscale systems?

Interconnected systems and processes:

- Single-cell behaviors
- Cell-cell communication
- Physics-imposed constraints (e.g., diffusion)
- Systems of systems (e.g., immune system)

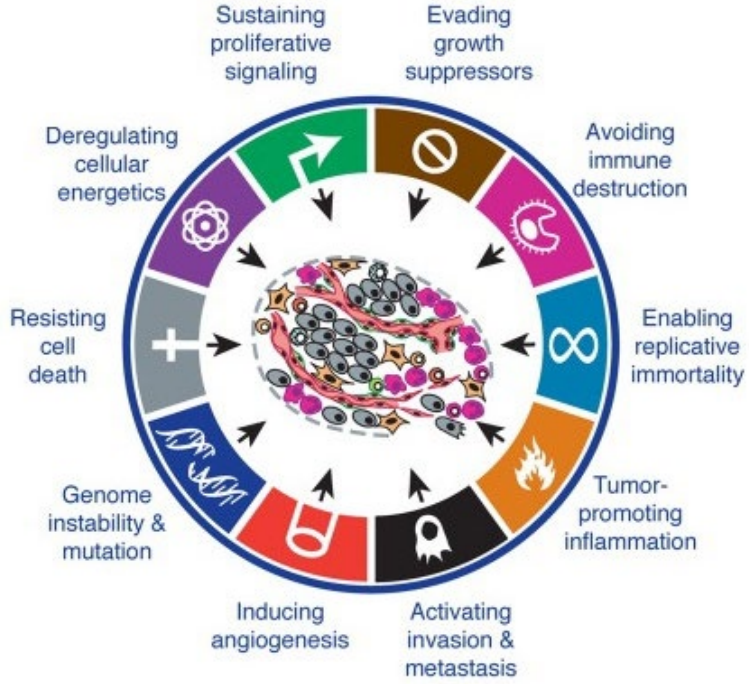
In diseases, these systems become dysregulated.

Treatments target *parts* of these systems.

Health is a **complex system:**
changing one part can have **surprising effects**!

Modeling can help **understand** this system.
This is **multicellular systems biology.**

If we can **control** these systems, we've arrived at **multicellular systems engineering**.



Source: Hanahan & Weinberg (2011)
DOI: 10.1016/j.cell.2011.02.013

Scientists use [models*] to detangle complex systems.

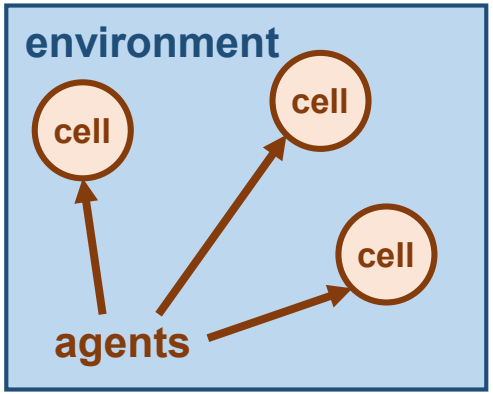
* animal, *in vitro*, engineered, mathematical, …

Key parts of a multicellular virtual laboratory

- **Model multiple diffusing chemical factors**
  - Growth substrates and metabolites
  - Signaling factors
  - Drugs
- **Model many cells in these chemical environments**
  - Environment-dependent behavior (including molecular-scale "logic")
  - Mechanical interactions
  - Heterogeneity:
    - individual states
    - individual parameter values
    - individual model rules
- **Run many copies of the model in high throughput**
  - Discover the rules that best match observations.
  - Identify and exploit weaknesses that can restore control

What is an agent-based model?

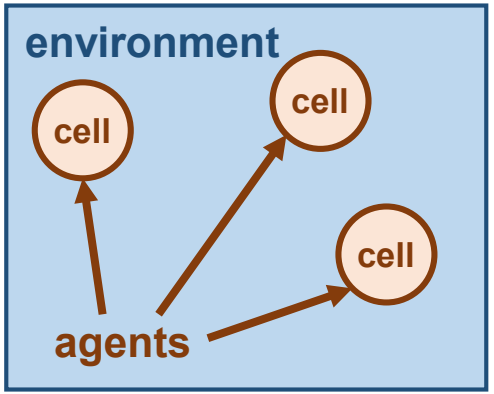
- Each cell is modeled as a separate software object (an **agent**) with:
  - **member data:** internal state variables
    - Position, Size, Cycle State, molecular variables, ….
  - **methods:** cellular processes
    - Cycling, Death, Motility, Growth, Adhesion, …



- Virtual cells move a virtual **(micro)environment**
  - Usually liquid (e.g., water or interstitial fluid)
  - Chemical movement (oxygen, glucose, signaling factors)
    - Typically diffusion: solve partial differential equations (PDEs)
    - May also require advection for environments with flow
  - May include mechanical components like extracellular matrix (ECM)
    - Finite element methods or related methods

What's the connection to biology and physics?

- The cell **agents encode our biological knowledge and hypotheses:**
  - Cell variables (member data) are selected to record important biological quantities
    - Volume, cell cycle state, energy, …
  - Cell rules (methods) encode biological hypotheses
    - Increase motility in low oxygen, down-regulate cycling under compression, …
  - Cell rules are often written at mathematical models
    - Potential functions for mechanics, systems of ODEs for metabolism, …



- The **microenvironment encodes physical constraints:**
  - *Chemical transport:* diffusion and advection equations (PDEs)
  - *Tissue mechanics:* viscoelastic, plastoelastic or other solid mechanics

- Most agent-based models combine **discrete** cell agents and **continuum** microenvironment processes. This is a **hybrid continuum-discrete approach.**

Simulation toolbox

BioFVM: Simulating 3-D biotransport

Design goal: Simulate multiple diffusing substrates in 3D with desktops or single HTC/HPC nodes

Typical use: pO_2, glucose, metabolic waste, signaling factors, and a drug, on 10 mm^3 at 20 μm resolution

Features:

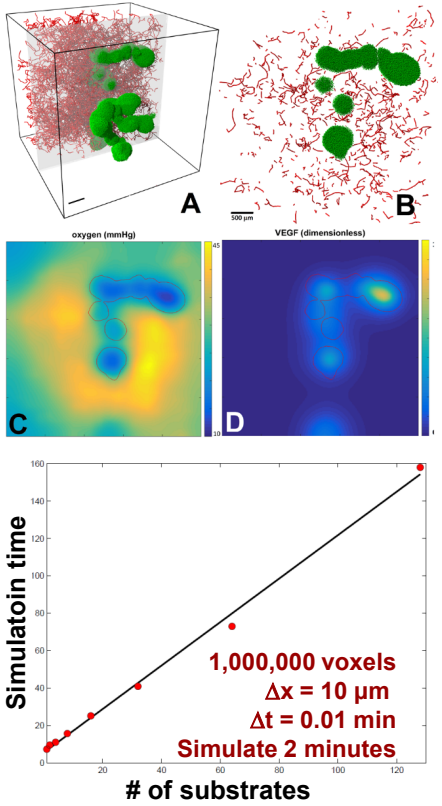
- Off-lattice cell secretion and uptake
- 2^{nd}-order accurate (space), 1^{st}-order accurate (time), numerically stable

Method:

- Operator splitting, LOD, customized Thomas solvers, etc.
- Standard C++11, cross-platform
- OpenMP parallelization
- O(*n*) cost scaling in # substrates, # voxels
- Easy to simulate 5-10 substrates on 10^6 voxels

Reference: Ghaffarizadeh et al., *Bioinformatics* (2016)

DOI: 10.1093/bioinformatics/btv730



PhysiCell: A multicellular framework

Design goal: Simulate 10^6 or more cells in 2D or 3D on desktops or single HPC nodes

Features:

- Off-lattice cell positions
- Mechanics-based cell movement
- Cell processes (cycling, motility, …)
- Signal-dependent phenotype
- Can dynamically attach custom data and functions on a cell-by-cell basis
- **Deployed from Raspberry Pi to Crays**

2019 PLoS Computational Biology Research Prize *for* Public Impact

Method:

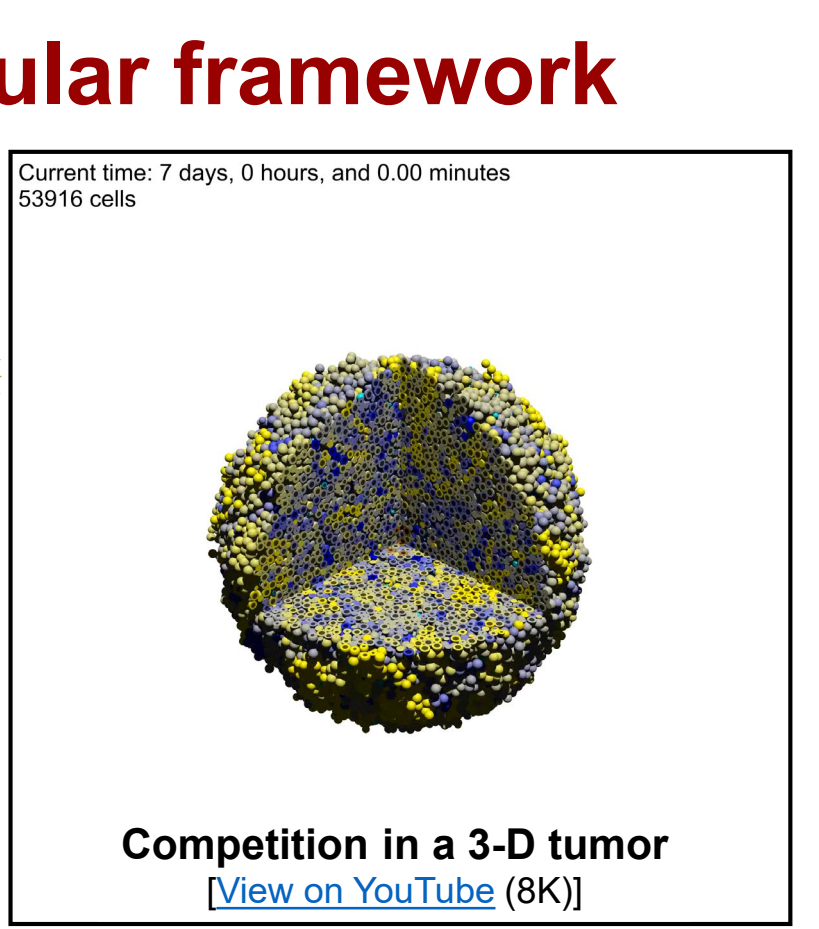
- Standard C++11, cross-platform
- OpenMP parallelization
- O(n) cost scaling in # cells

Reference: Ghaffarizadeh et al., PLoS Comput. Biol. (2018)

DOI: 10.1371/journal.pcbi.1005991

Try this model yourself!

nanohub.org/tools/pc4heterogen



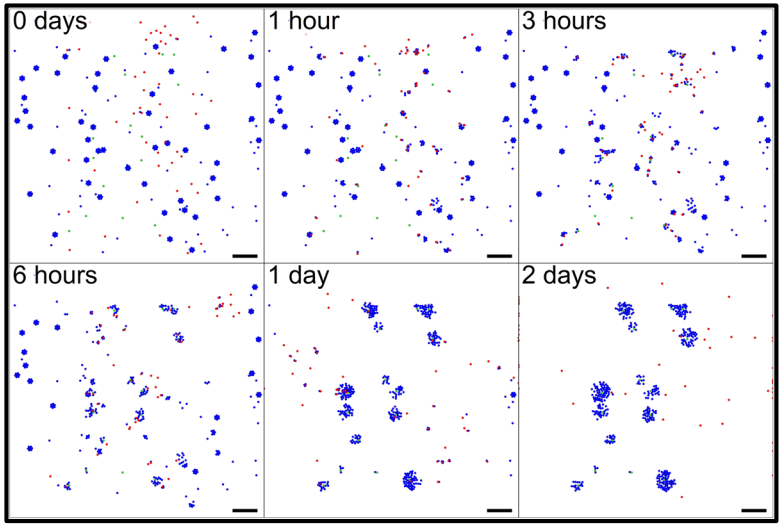
Competition in a 3-D tumor

[View on YouTube (8K)]

Let's try a model!

Example: biological cargo delivery system

- **Chemical environment:**
  - two diffusing chemical signals
- **Cell types and rules:**
  - **directors (green):**
    - secrete director signal to attract workers
  - **cargo (blue):**
    - **undocked**: secrete cargo signal to attract workers
    - **docked**: turn off signal
  - **workers (red):**
    - **undocked**: seek cargo via chemotaxis
    - **docked:** seek directors via chemotaxis, release cargo in high signal areas



Try this model yourself!

https://nanohub.org/tools/pc4biorobots

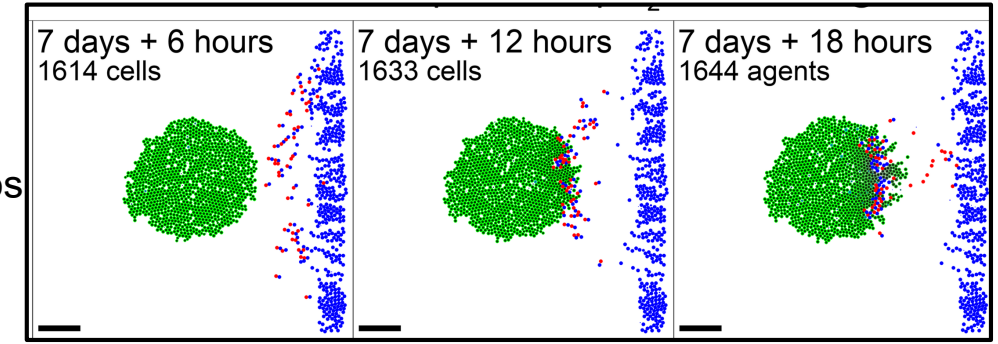
pc4biorobots exercises

1. **Cargo and workers only**
   - Set # of directors to zero.
   - Set max time to 120 minutes.
   - Click run. What happens?
   - Plot the cargo signal. How does this explain the behavior?
2. **Full model**
   - Set # of directors to 15
   - Set max time to 1000 minutes.
   - Click run. What happens?
   - Plot the director signal. How does this explain the behavior?
3. **Modify workers (1)**
   - Set drop threshold to 0.1
   - Click run. What happens?
   - Plot the director signal. How does this explain the behavior?
4. **Modify workers (2)**
   - Set attached migration bias to 0.3.
   - Click run. What happens?

What if we could use this for cancer treatments?

pc4cancerbots

- **cancer biorobots:**
  - **green:**
    - cycle entry scales with O2
    - O2 depletion causes necrosis
    - cumulative drug exposure causes apoptos
  - **blue:**
    - drug-loaded "cargo"
  - **red:**
    - worker cells that seek and haul cargo towards hypoxic zones



Try this model yourself!

https://nanohub.org/tools/pc4cancerbots

pc4cancerbots exercises (later)

1. **Cancer cells only**
   - Set # injected cells to 0
   - Increase tumor radius to 400
   - Set max time to 2880 minutes.
   - Click run. What happens?
   - Plot the oxygen. How does this explain the behavior?
2. **Add therapy (full model)**
   - Set # of injected cells to 500
   - Set therapy activation time to 120
   - Increase max time to 4320 minutes
   - Click run. What happens?
   - Plot the therapeutic. How does this explain the behavior?
3. **Modify treatment**
   - Set attached worker migration bias to 0.2
   - Click run. What happens?
4. **Modify treatment (on your own)**
   - Set cargo release o2 threshold to 15
   - Increase max time to 14400 minutes
   - Click run. What happens?

This is a key strength of simulation models:

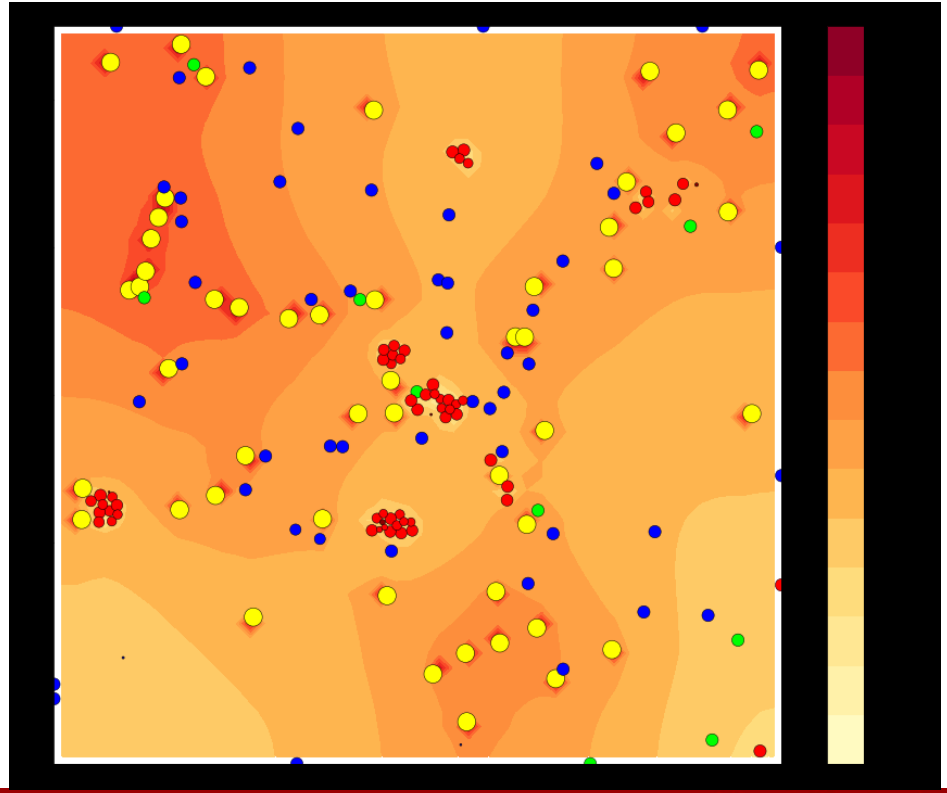
We can explore new ideas before committing time and resources.

pcISA: an adversarial multicellular system

- What are the dynamics of an adversarial system?
- **suppliers (e.g., blood vessels)**
  - supply growth substrates
- **Invaders (e.g., bacteria)**
  - grow near vessels
  - avoid dead cells
  - avoid attackers
- **Scouts (e.g., macrophages)**
  - look for invaders, release signal
- **Attackers (e.g., T cells)**
  - Look for signal, attack invaders

Try this model yourself!

https://nanohub.org/tools/pcisa



pcISA exercises

1. **Suppliers and invaders only**
   - Set # of scouts and # of attackers to 0
   - Set max time to 2400 minutes.
   - Click run. What happens?
   - Plot the resource. How does this explain the behavior?
2. **Add scouts**
   - Set # of scouts to 10
   - Click run. What happens? Does plotting the "signal" help explain their behavior?
3. **Add attackers (full model)**
   - Set # of attackers to 50
   - Set max time to 7200 minutes.
   - Click run. What happens?
4. **Modify invaders**
   - Set invader quorum weight to 0.01
   - Click run. What happens?
   - Plot the death signal. How does this explain the behavior of invaders after an attack?
5. **Modify invaders and scouts (on your own)**
   - Set invader quorum weight to 1
   - Set scout migration bias to 1
   - Increase invader max death rate to 0.01
   - Click run. What happens?

3-Types model

- In physics, the ***3-body problem*** shows how 3 objects with very simple interactions (gravitation) can demonstrate chaotic behavior.
- **Let's build a similar system for biology!**

- **3 cell types** (**A**,**B**,**C**) each secrete their own chemical factor

- Each cell type can:
  - **divide** and **die** in response to resource (R), A, B, C, and pressure
  - **move** in response to A, B, C, and R
  - **secrete** (or not secrete) in response to A, B, C, and R

- ***What can happen in this general system?***

Try this model yourself!

https://nanohub.org/tools/pc3types

pc3types exercises (competitive)

1. **Competition for resources (neutral)**
   - Increase to 75 of A, B, and C cells
   - Decrease max time to 5760 minutes (4 days)
   - Run with default parameters – what happens?
2. **Competition for resources (each type secretes "poisons")**
   - For type A: B and C promote death (apoptotic death rate 0.001)
   - For type B: A and C promote death (apoptotic death rate 0.001)
   - For type C: A and B promote death (apoptotic death rate 0.001)
3. **Competition for resources (A more aggressive)**
   - Change Type A's Phase 0->Phase 1 transition rate to 0.005
4. **Competition for resources (A more aggressive, B is motile)**
   - Change Type B's motility to "on", migration bias = 0.5, towards "resource"

pc3types exercises (cooperative)

1. **A helps B, and B helps C**
   - Reset to defaults
   - Decrease max time to 5760 minutes (4 days)
   - Type B: A promotes division
   - Type C: B promotes division
2. **A helps B, B migrates to A. B helps C.**
   - Type B: A promotes division, chemotaxis towards A
   - Type C: B promote division
3. **A helps B, B migrates to A. B helps C. (Version 2)**
   - Type B: A promotes division, chemotaxis towards A. A inhibits migration
   - Type C: B promote division
4. **A helps B, B migrates to A. B suppresses proliferation of A.**
   - Type B: A promotes division, chemotaxis towards A. A inhibits migration
   - Type A: B inhibits division
   - Type C: B promotes division. chemotaxis away from B. B promotes migration.
   - Set diffusion coefficient of factors A, B, C to 100, set decay to 0.04 (length scale = 50 micron)

pc3types exercises (repressilator)

1. Set A, B, and C diffusion coefficients to 100
2. Set A, B, and C decay rates to 0.01
3. Type A:
   - Use 500 cells, all within 400 microns
   - C inhibits secretion
   - No birth or death
4. Type B:
   - Use 500 cells, all within 400 microns
   - A inhibits secretion
   - No birth or death
5. Type C:
   - Use 500 cells, all within 400 microns
   - B inhibits secretion
   - no birth or death

Further reading (1)

- **BioFVM method paper (3-D diffusion)**
  A. Ghaffarizadeh, S.H. Friedman, and P. Macklin. BioFVM: an efficient, parallelized diffusive transport solver for 3-D biological simulations. *Bioinformatics* 32(8):1256-8, 2016. DOI: 10.1093/bioinformatics/btv730.

- **PhysiCell method paper (agent-based model)**
  A. Ghaffarizadeh, R. Heiland, S.H. Friedman, S.M. Mumenthaler, and P. Macklin. PhysiCell: an open source physics-based cell simulator for 3-D multicellular systems. *PLoS Comput. Biol.* 14(2):e1005991, 2018. DOI: 10.1371/journal.pcbi.1005991.

- **PhysiBoSS (PhysiCell + MaBoSS for Boolean networks)**
  G. Letort, A. Montagud, G. Stoll, R. Heiland, E. Barillot, P. Macklin, A. Zinovyev, and L. Calzone. PhysiBoSS: a multi-scale agent based modelling framework integrating physical dimension and cell signalling. *Bioinformatics* 35(7):1188-96, 2019. DOI: 10.1093/bioinformatics/bty766.

- **xml2jupyter paper (create GUIs for cloud-hosted models)**
  R. Heiland, D. Mishler, T. Zhang, E. Bower, and P. Macklin. xml2jupyter: Mapping parameters between XML and Jupyter widgets. *Journal of Open Source Software* 4(39):1408, 2019. DOI: 10.21105/joss.01408.

- **PhysiCell+EMEWS (high-throughput 3D PhysiCell investigation)**
  J. Ozik, N. Collier, J. Wozniak, C. Macal, C. Cockrell, S.H. Friedman, A. Ghaffarizadeh, R. Heiland, G. An, and P. Macklin. High-throughput cancer hypothesis testing with an integrated PhysiCell-EMEWS workflow. *BMC Bioinformatics* 19:483, 2018. DOI: 10.1186/s12859-018-2510-x.

- **PhysiCell+EMEWS 2 (HPC accelerated by machine learning)**
  J. Ozik, N. Collier, R. Heiland, G. An, and P. Macklin. Learning-accelerated Discovery of Immune-Tumour Interactions. *Molec. Syst. Design Eng.* 4:747-60, 2019. DOI: 10.1039/c9me00036d.

Further reading (2)

- **A review of cell-based modeling (in cancer):**

  J. Metzcar, Y. Wang, R. Heiland, and P. Macklin. A review of cell-based computational modeling in cancer biology. *JCO Clinical Cancer Informatics* 3:1-13, 2019 (invited review). DOI: 10.1200/CCI.18.00069.

- **Progress on multicellular systems biology:**

  P. Macklin, H.B. Frieboes, J.L. Sparks, A. Ghaffarizadeh, S.H. Friedman, E.F. Juarez, E. Jockheere, and S.M. Mumenthaler. "Progress Towards Computational 3-D Multicellular Systems Biology". In: . Rejniak (ed.), *Systems Biology of Tumor Microenvironment*, chap. 12, pp. 225-46, Springer, 2016. ISBN: 978-3-319-42021-9. (invited author: P. Macklin). DOI: 10.1007/978-3-319-42023-3_12.

- **Challenges for data-driven multicellular systems biology**

  P. Macklin. Key challenges facing data-driven multicellular systems biology. *GigaScience* 8(10):giz127, 2019. DOI: 10.1093/gigascience/giz127

Some models to explore

On nanoHUB:

- **pc4heterogen:** heterogeneous cancer growth (https://nanohub.org/tools/pc4heterogen)
- **pc4cancerbots:** use the "biorobots" as a cell-based cancer therapy (https://nanohub.org/tools/pc4cancerbots)
- **pc4cancerimmune:** basic cancer immunotherapy model (https://nanohub.org/tools/pc4cancerimmune)
- **trmotility**: learn about biased random cell migration (https://nanohub.org/tools/trmotility)
- **pcisa:** learn about an adversarial ecosystem: invader cells are fueled by resource providers, but scout cells seek invaders to recruit attackers, who poison invaders. (https://nanohub.org/tools/pcisa)
- **pc4thanos:** *Avengers Endgame* battle using cell rules (https://nanohub.org/tools/pc4thanos)
- **pc4covid19:** COVID-19 simulation model (https://nanohub.org/tools/pc4covid19)
- **pc4livermedium:** tumor-stroma biomechanical feedbacks (https://nanohub.org/tools/pc4livermedium)