Design principles of complex cell-fate decision networks: Examples in development & cancer – *Part 2*

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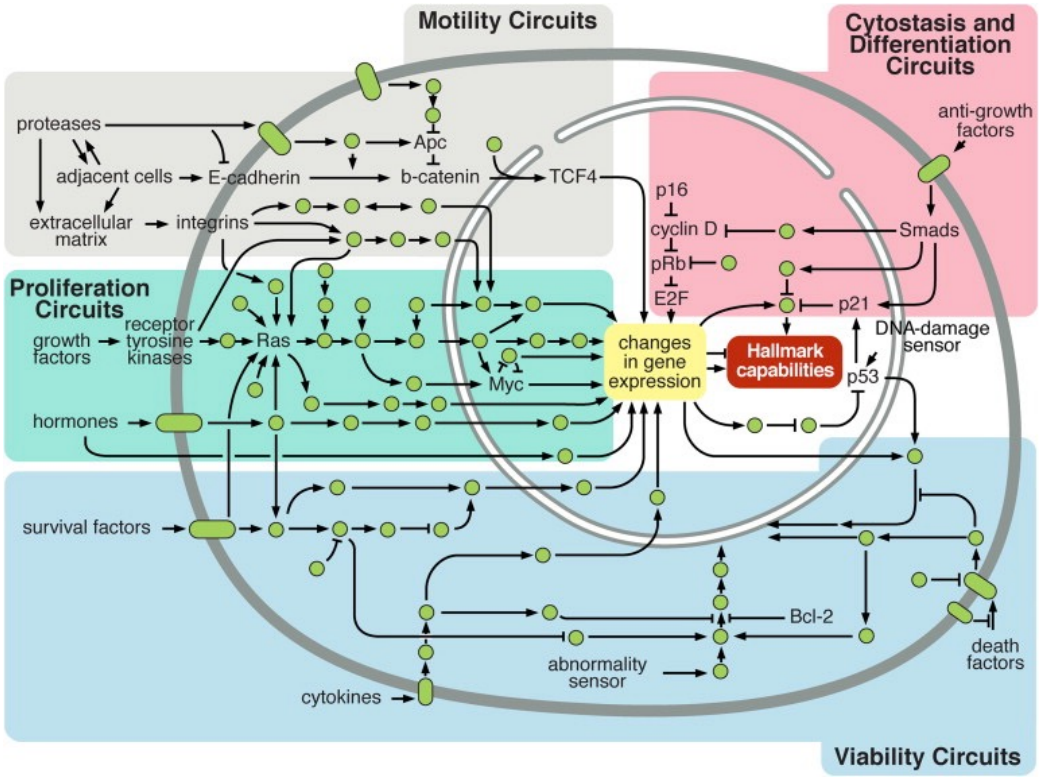
Editor-in-Chief, NPJ Systems Biology & Applications

Workshop on Flags, Landscapes and Signals | IMSc Chennai

mkjolly15

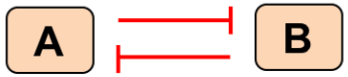
Cellular decision-making



- Cells receive diverse biophysical/chemical signals varying in (x, t).
- Cells in a population can respond differently to the same signals.
- Cellular decision-making is driven by interconnected complex networks.

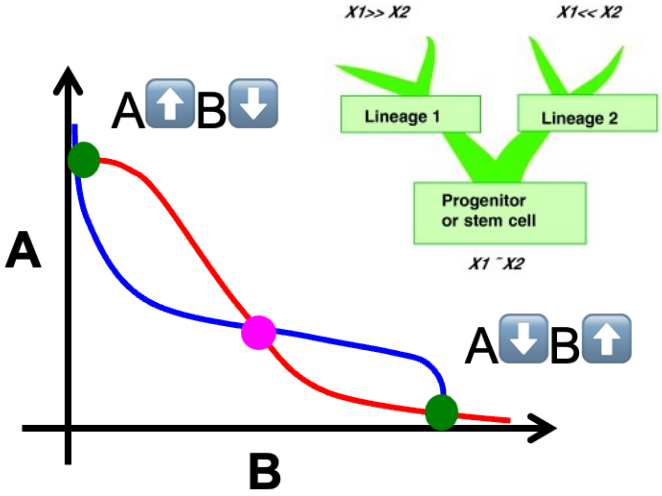
Hanahan & Weinberg, Cell 2011

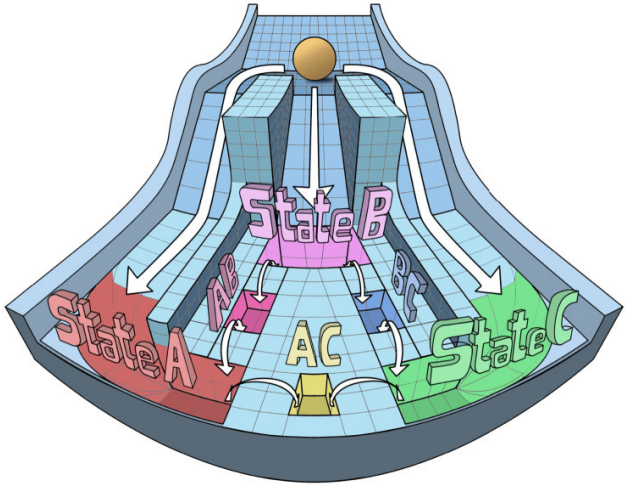
Summary from Part 1



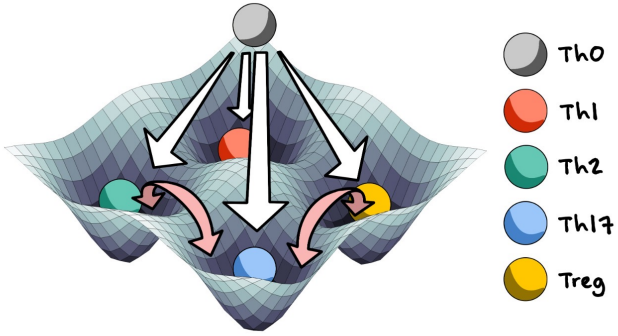
Bistability
(A, B) = (high, low)
(A, B) = (low, high)

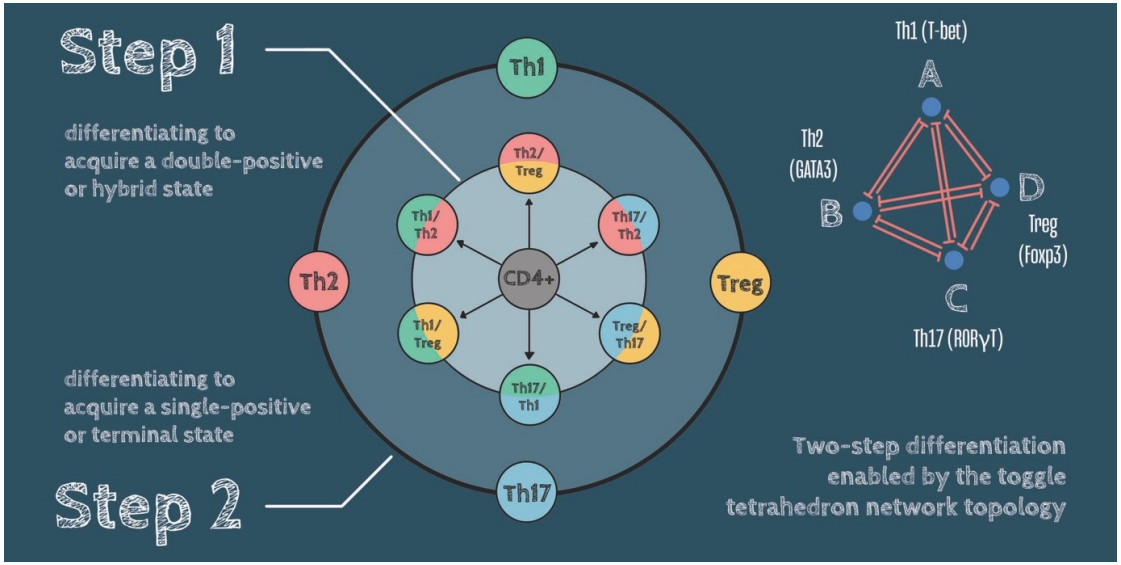
Huang, PloS Biology 2013
Gardner *et al.* Nature 2000





CD4+ T-cell differentiation



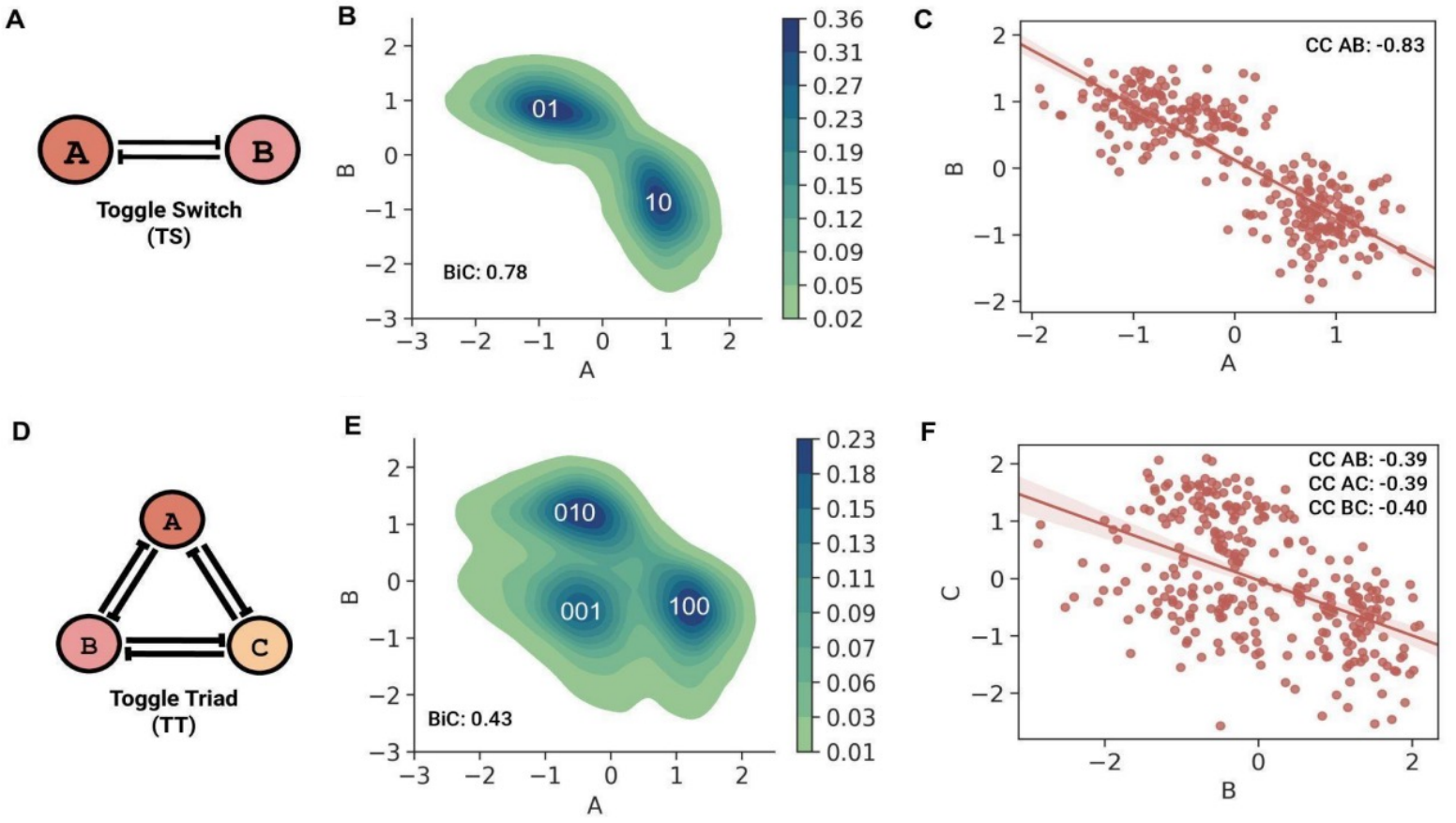


Outline for today

- Impact of embedding 2-node, 3-node network motifs in larger networks

- Investigating larger gene regulatory networks to identify similarities with 2-node, 3-node network motifs

(Stand-alone) Dynamics of toggle switch, triad



How does this dynamics change when embedded in large networks?

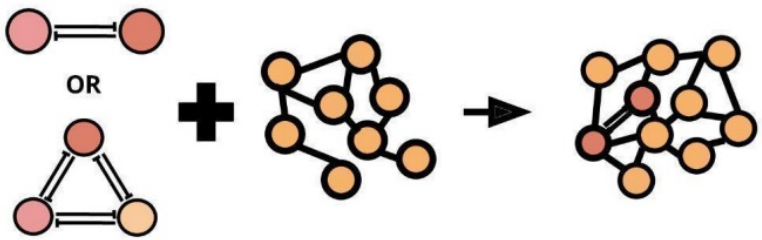
Embedding TS, TT in an ensemble of large networks

A

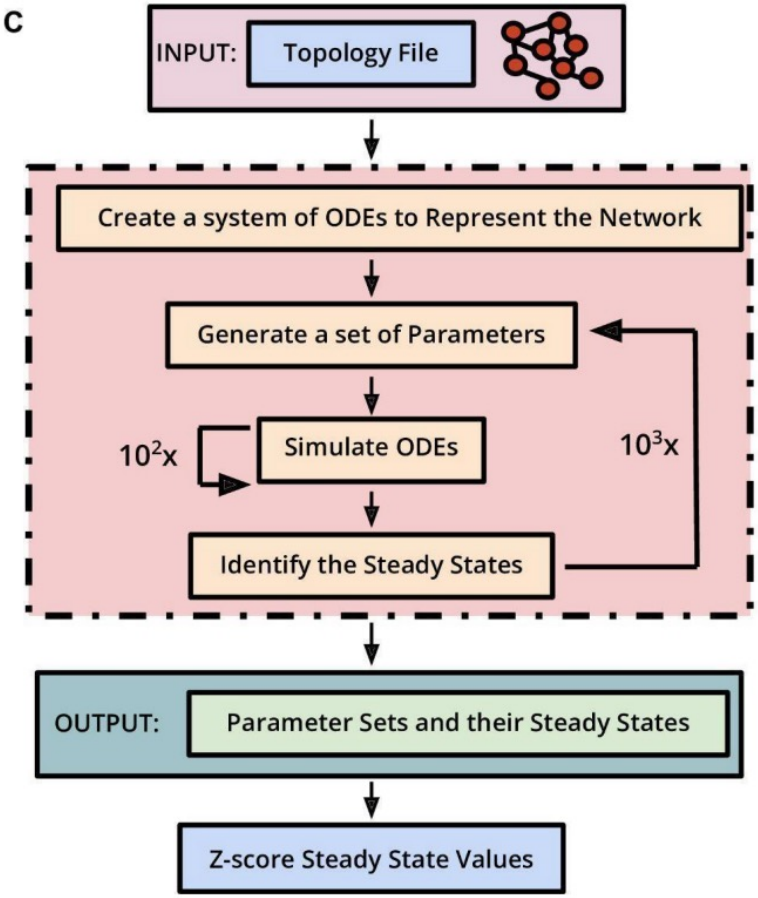
Order	Mean Connectivity		
	E:2N	E:4N	E:6N
5N	5N | E:2N	5N | E:4N	5N | E:6N
10N	10N | E:2N	10N | E:4N	10N | E:6N
15N	15N | E:2N	15N | E:4N	15N | E:6N
20N	20N | E:2N	20N | E:4N	20N | E:6N

B

Embedding TS or TT motif into larger networks:



C



Pradyumna

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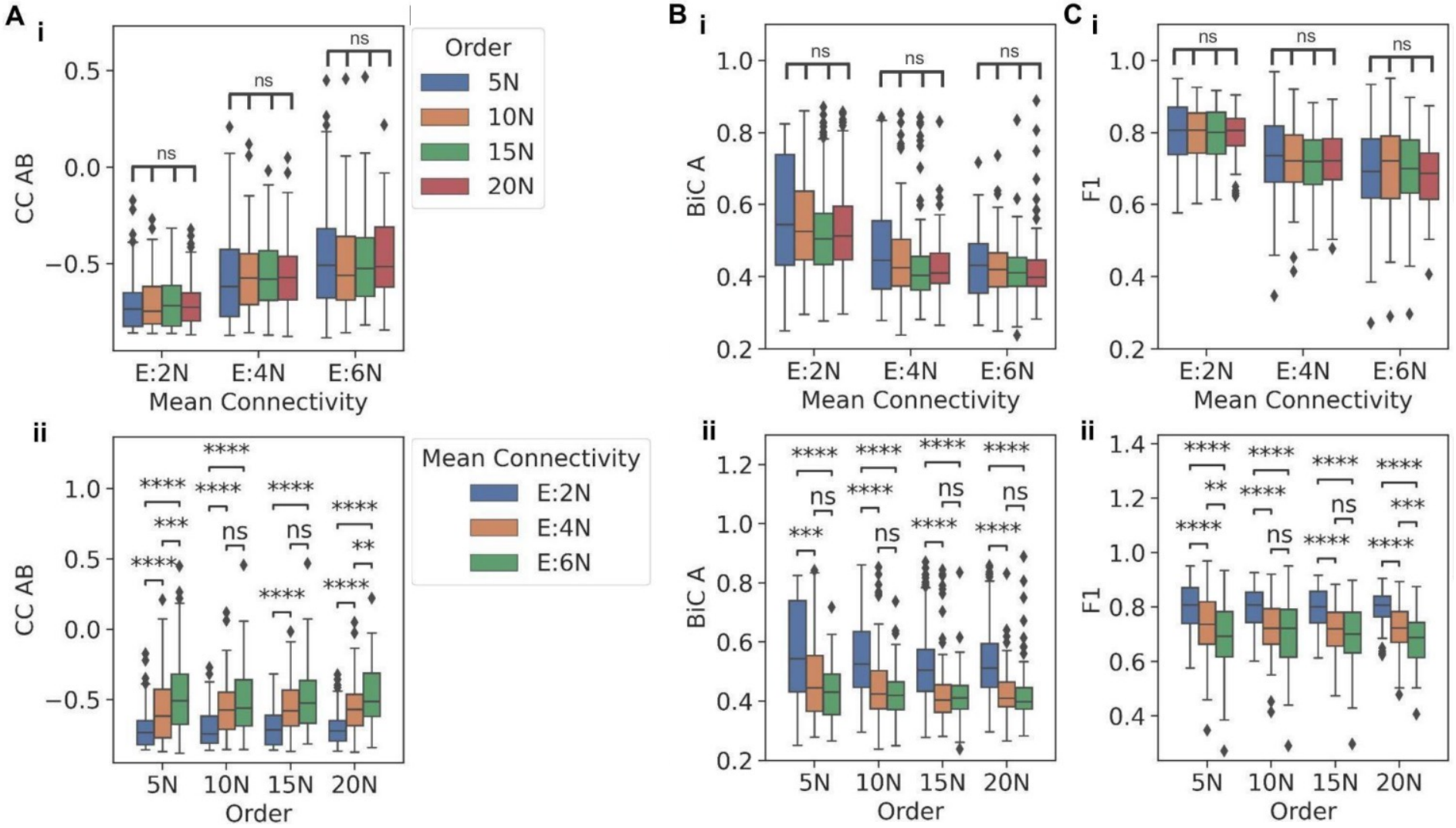
Global density impacts the dynamics of embedded TS



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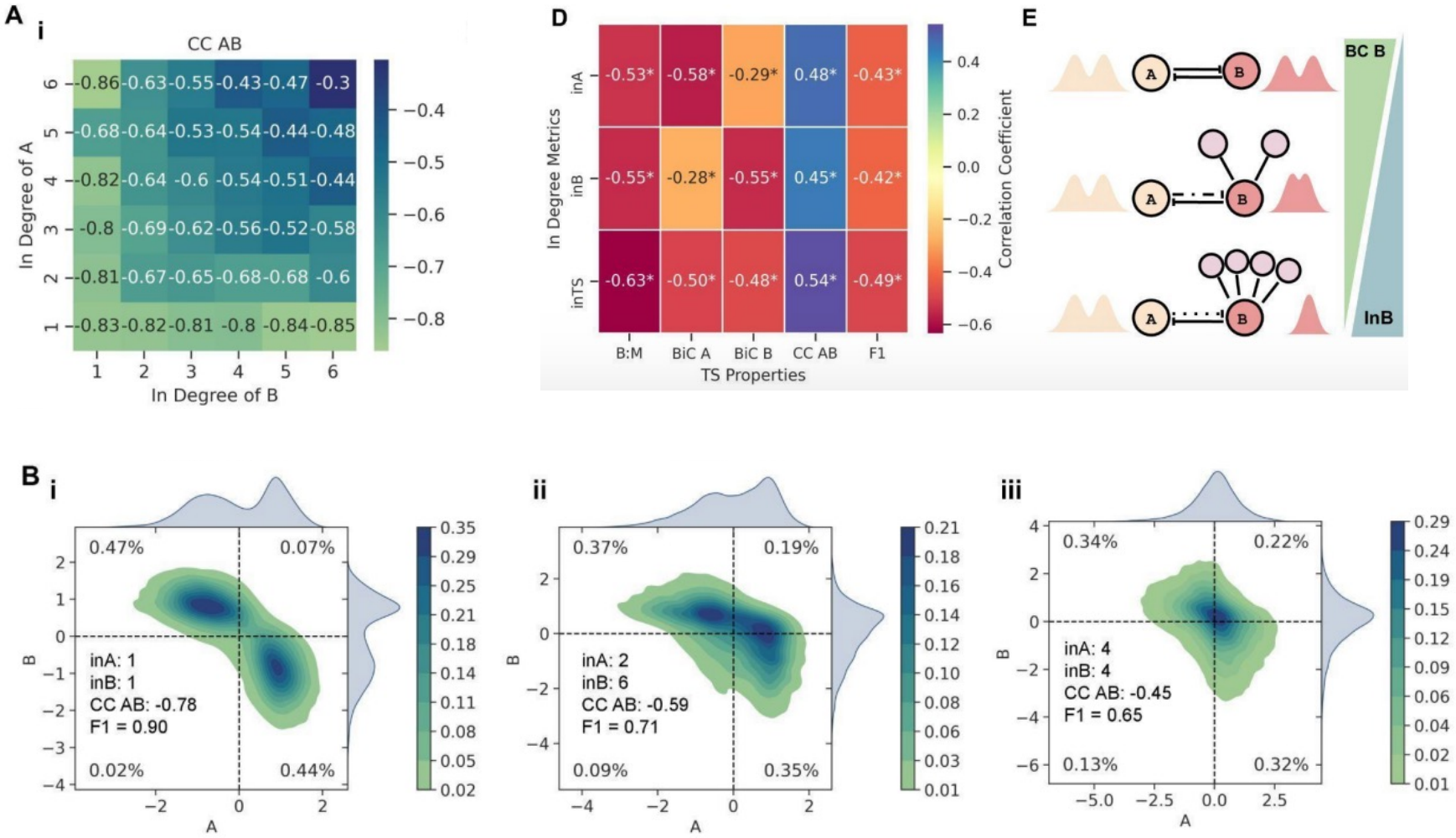
Local density around a TS influences its dynamics



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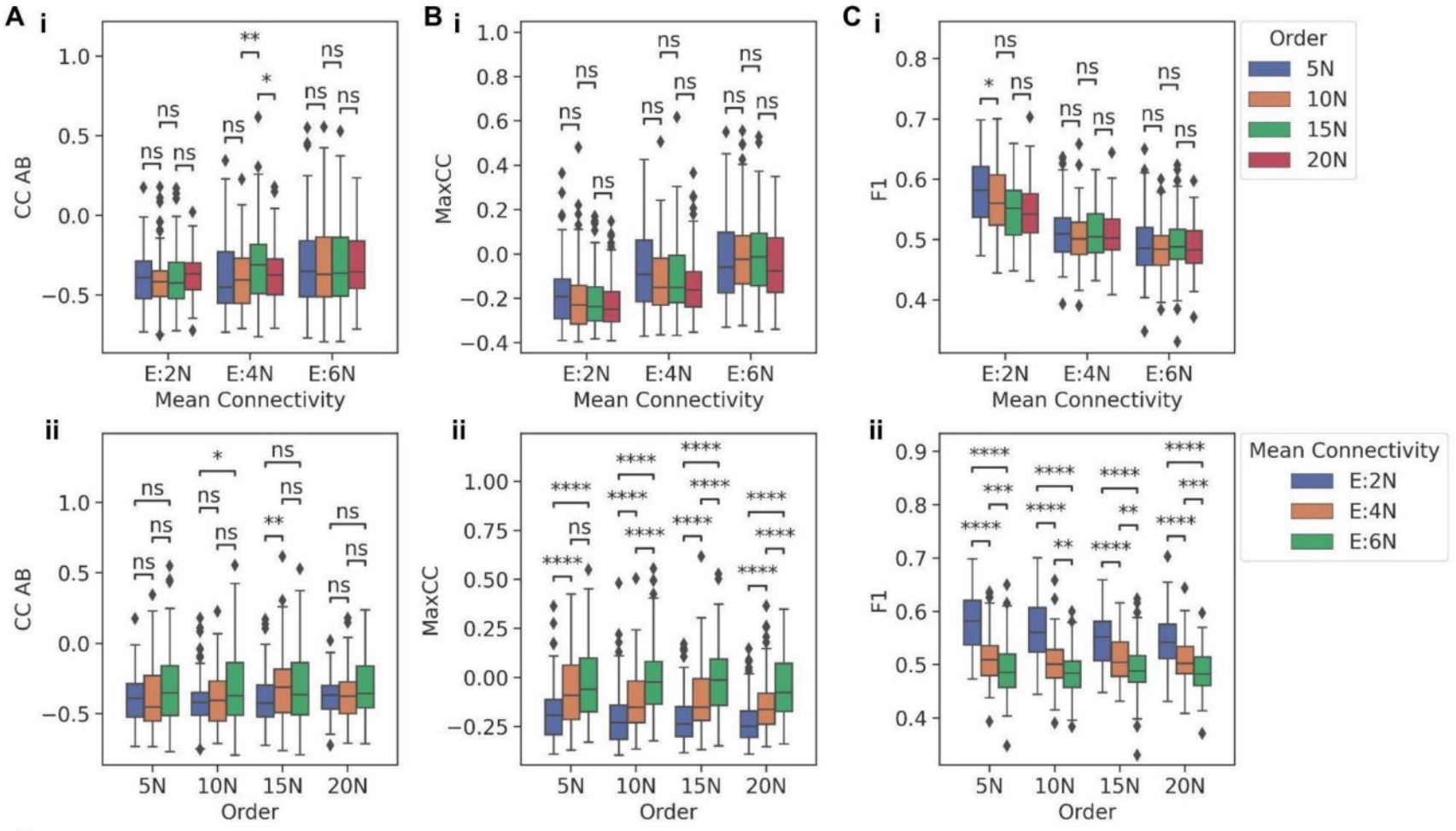
Global density impacts the dynamics of embedded TT



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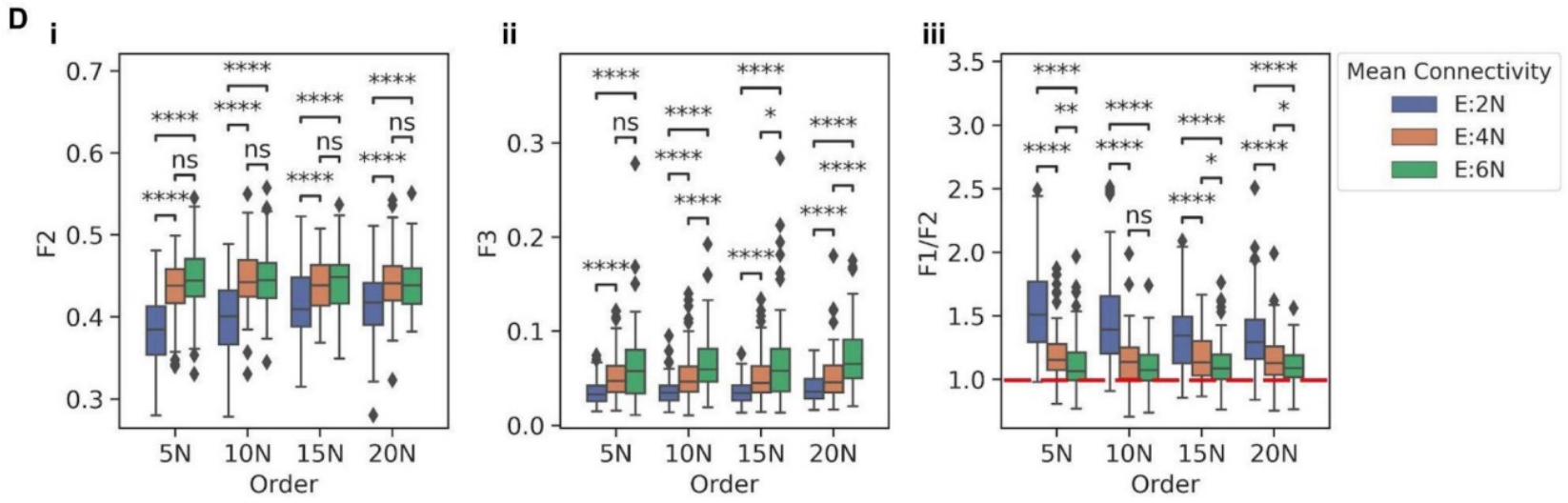
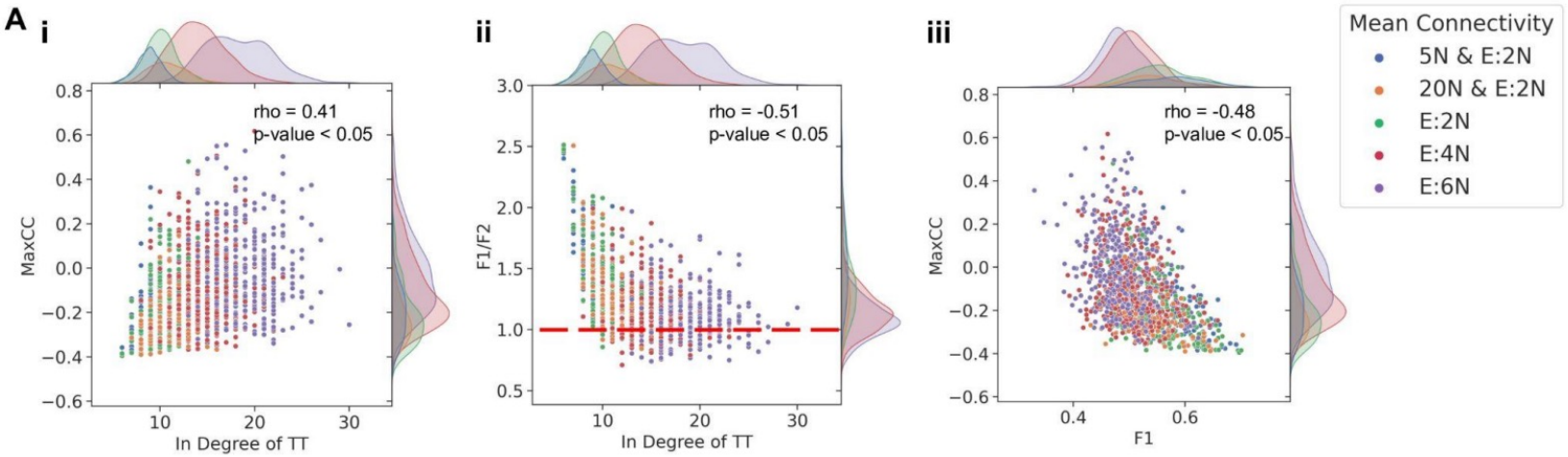
Embedded TT get enriched for ‘double positive’ states





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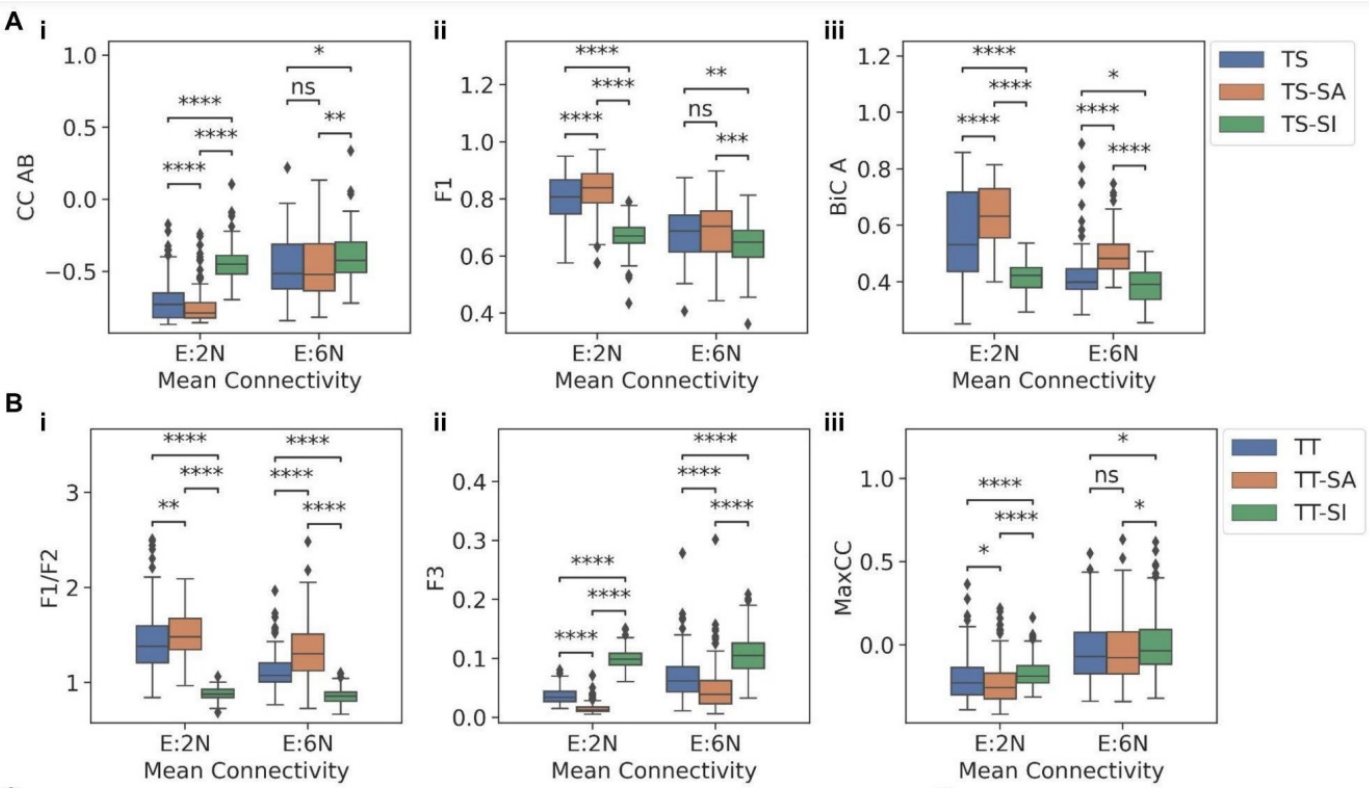
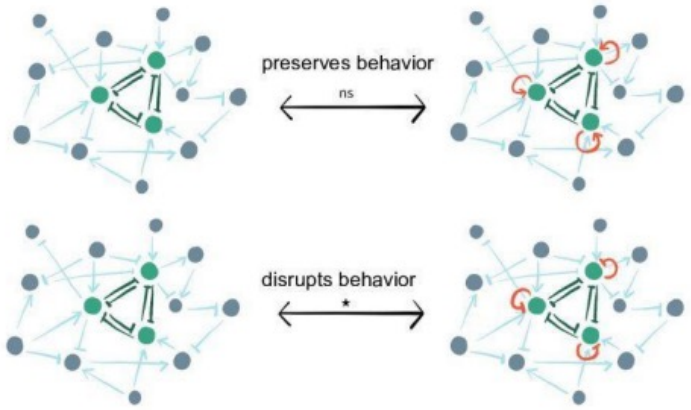
Impact of self-regulation on embedded TS, TT dynamics





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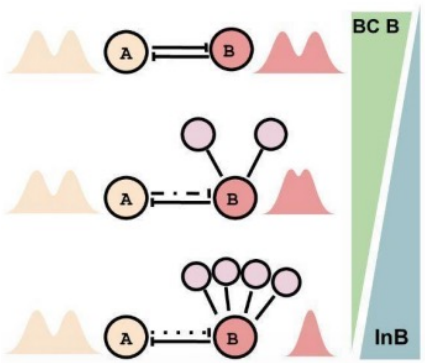
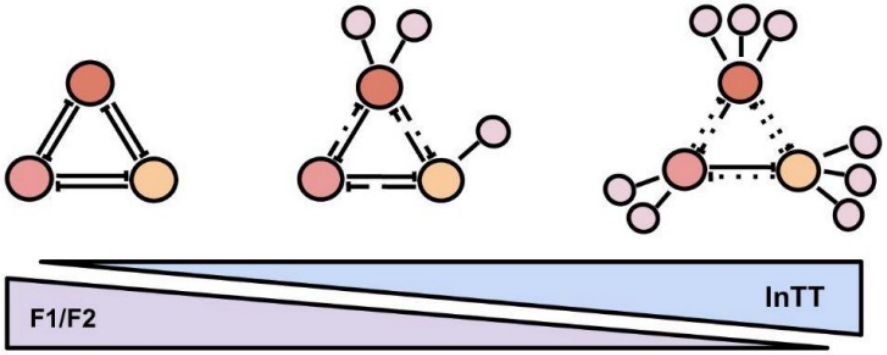
Summary so far

How do these network motifs operate when embedded in larger networks?



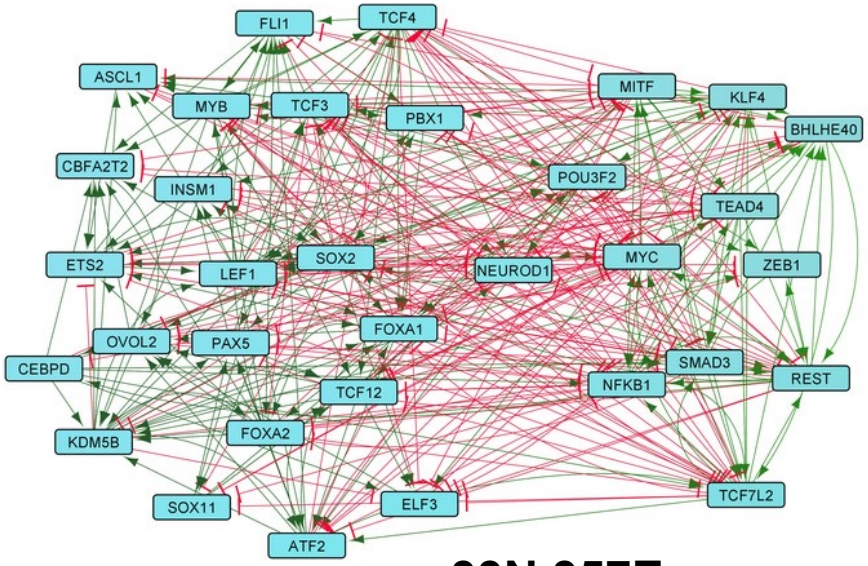


High local density (irrespective of sign of incoming edges) around a TS or TT and self-inhibition on TS, TT nodes disrupt the stand-alone features of TS, TT.

Note: *E.coli* transcriptional network has average in-degree < 3.

Regulatory network for small cell lung cancer (SCLC)

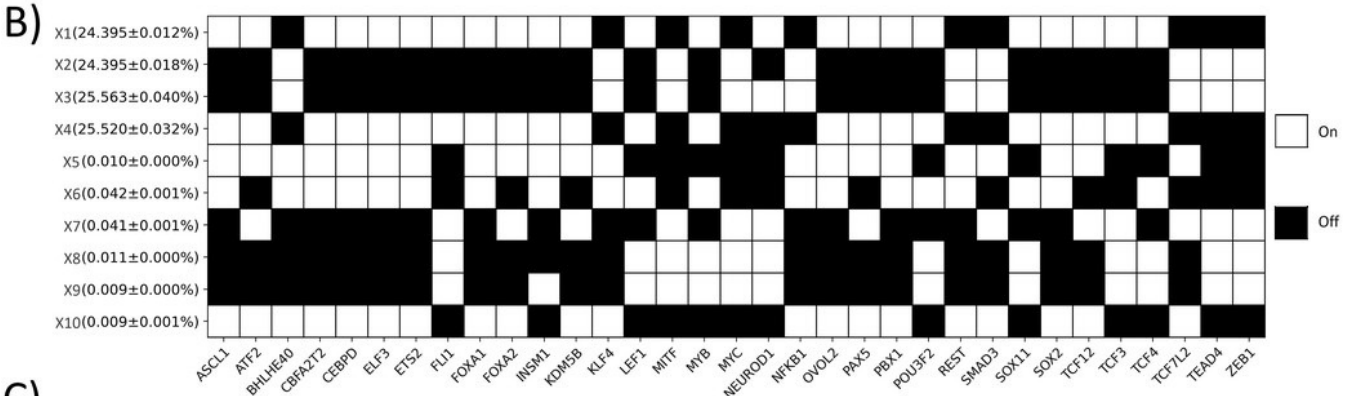
A)



33N 357E

$$s_i(t+1) = \begin{cases} +1, \sum_j Adj_{ij} s_j(t) > 0 \\ -1, \sum_j Adj_{ij} s_j(t) < 0 \\ s_i(t), \sum_j Adj_{ij} s_j(t) = 0 \end{cases}$$

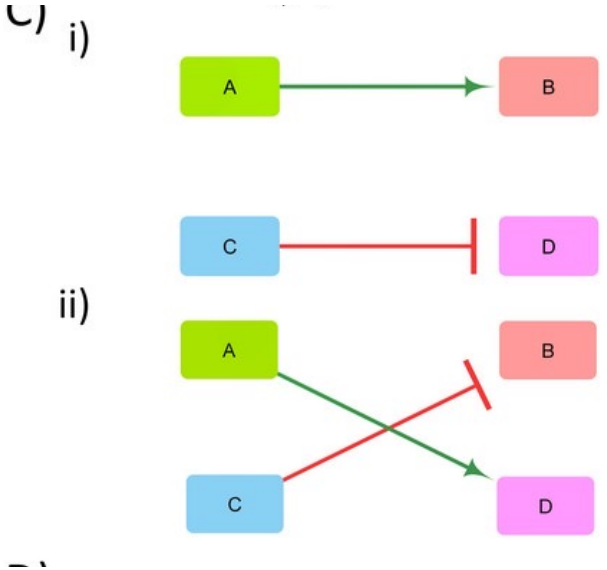
B)

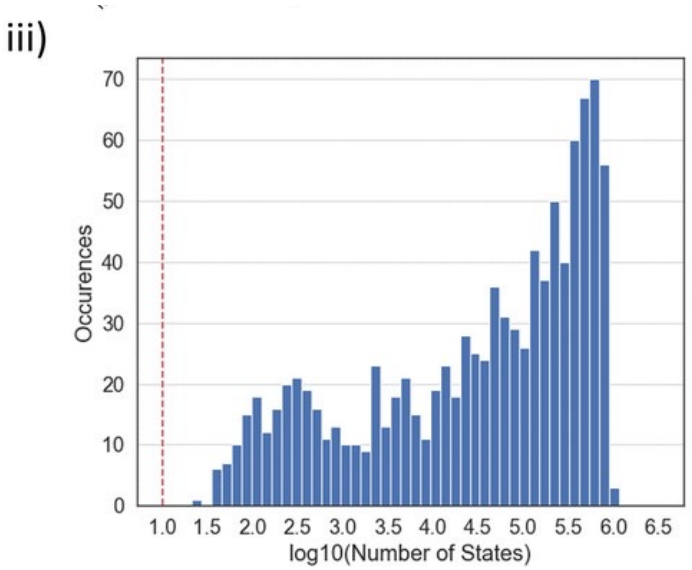


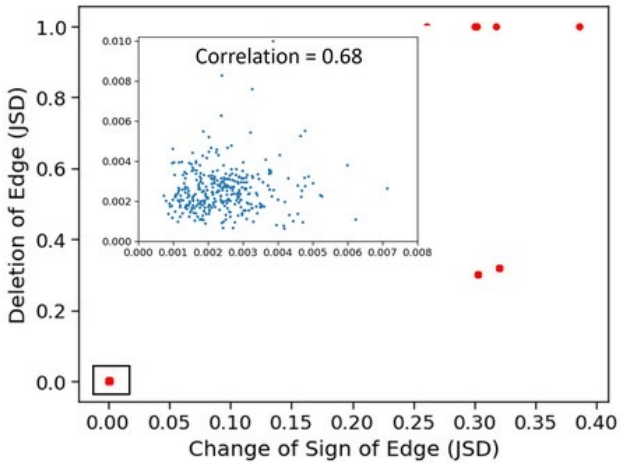
C)

Chauhan *et al.* eLife 2021

Regulatory network for small cell lung cancer (SCLC)





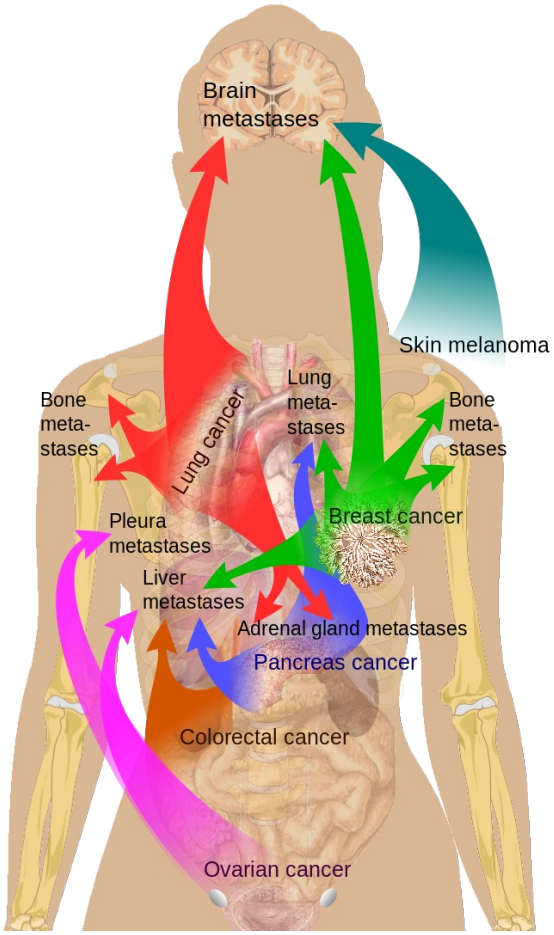


In only 12 out of 357 cases, a significant change in steady-state distribution was noted.

=> Very high robustness

Chauhan *et al.* eLife 2021

Metastasis : the cause of 90% of all cancer deaths



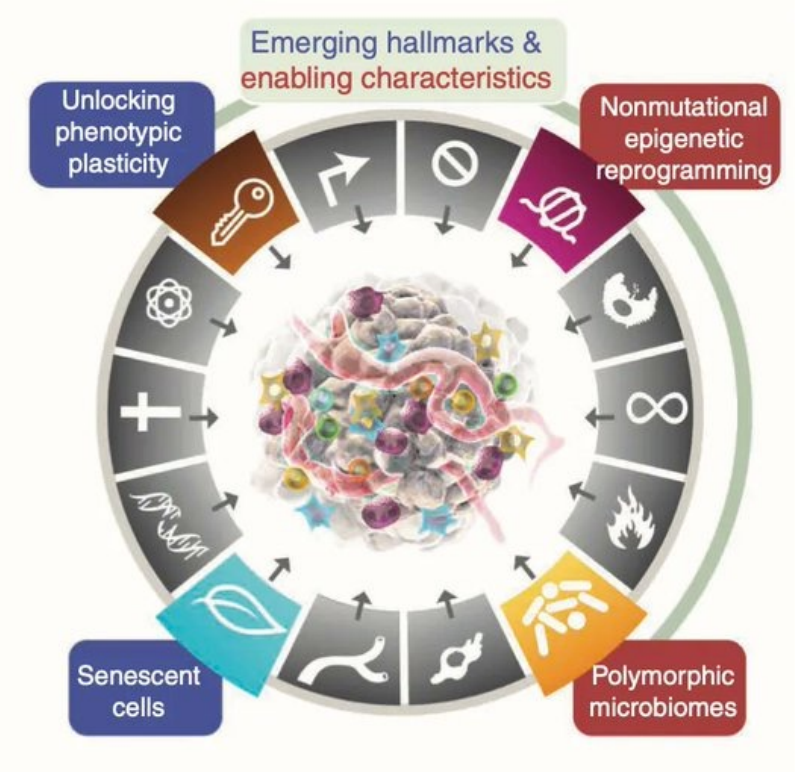
No unique mutational signatures yet identified for metastasis.

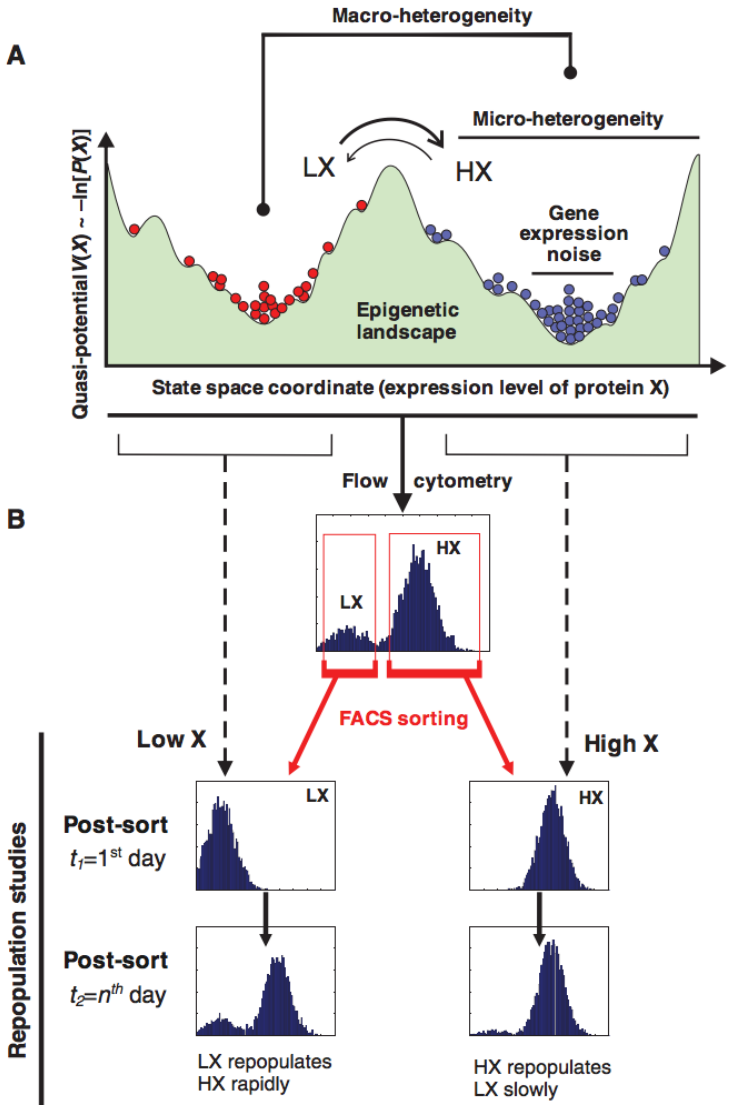
Metastasis is a highly inefficient process (<0.02%)

Phenotypic plasticity : a hallmark of metastasis

Phenotypic plasticity:

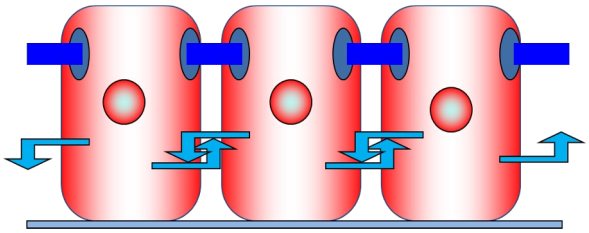
- Ability of cells to switch their cell-state reversibly in response to environmental conditions
- Fast, reversible (unlike mutations)



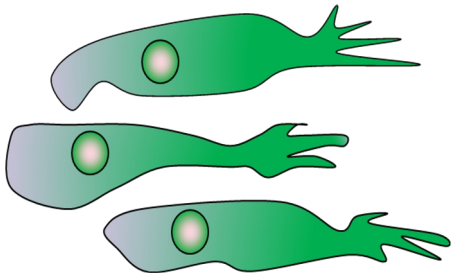


Huang *et al.* Development 2009
Hanahan, Cancer Discov 2022

EMT/MET: The engine of metastasis

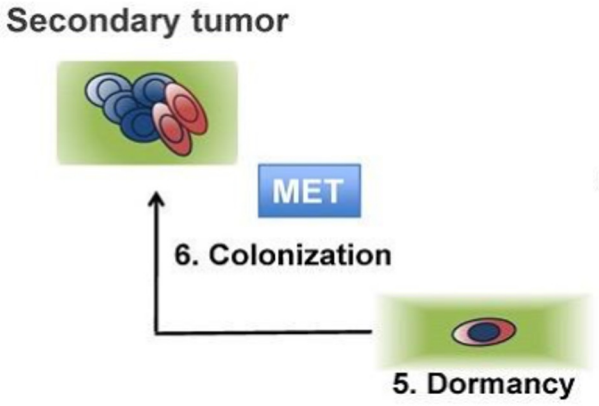


Adhere to neighbors
Do NOT migrate or invade
Epithelial (E)

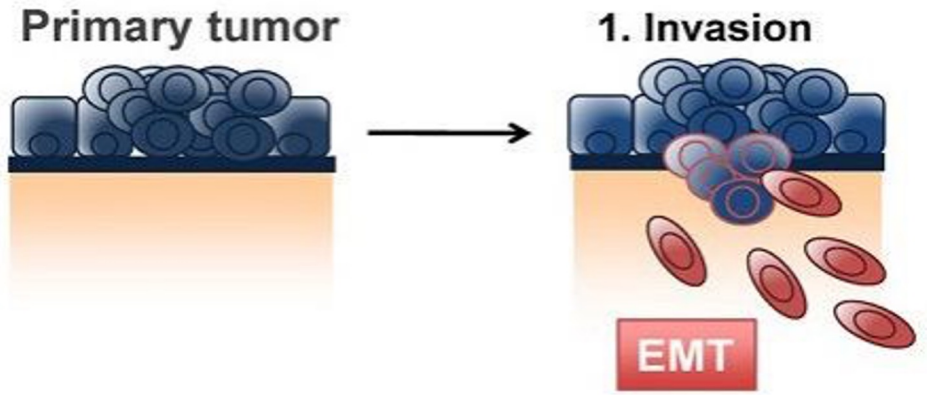


Do NOT adhere to neighbors
Migrate and invade
Mesenchymal (M)

Mesenchymal-to-Epithelial Transition (MET)

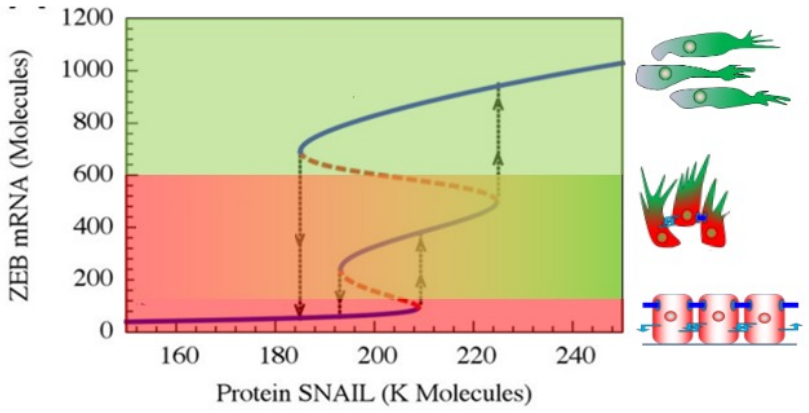


Epithelial-to-Mesenchymal Transition (EMT)

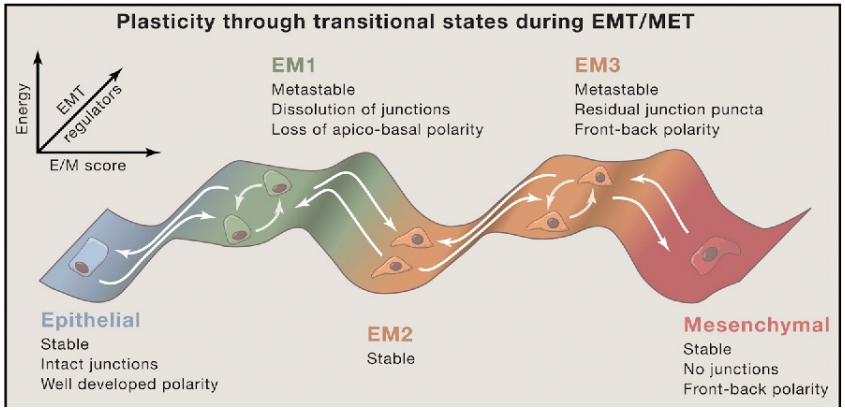


Scheel & Weinberg, Semin Cancer Bio 2012

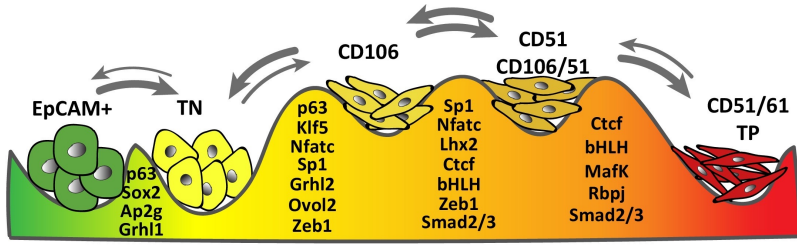
From EMT (2002-2012) to EMP (2013-now)



Lu#, Jolly# *et al.* PNAS 2013

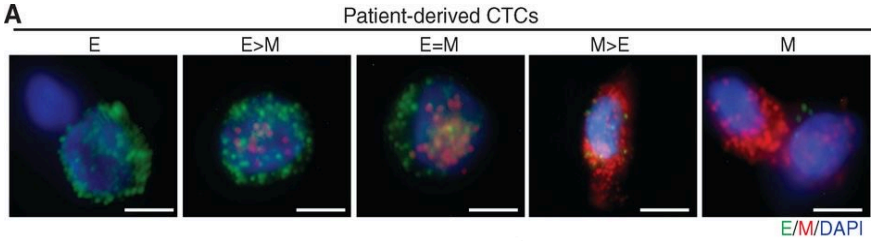


Nieto *et al.* Cell 2016



	Epithelial tumor cells	Early hybrid EMT state	Hybrid EMT state	Late hybrid EMT state	Mesenchymal tumor cells
Proliferation	+++++	++++	+++	++	+
Invasion	+	++	+++	++++	+++++
Plasticity	+	++	+++	++++	++
Stemness	+	+++	+++	+++	+++
Metastasis	+	++++	++++	++	+

Pastushenko & Blanpain, Trends Cell Biol 2019
Pastushenko *et al.* Nature 2018

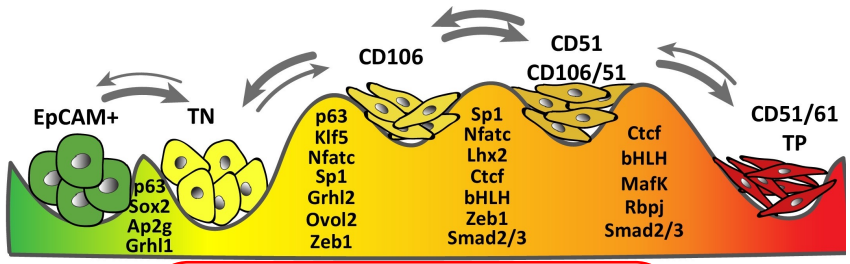


Acquisition of a hybrid E/M state is essential for tumorigenicity of basal breast cancer cells

Cornelia Kröger[a], Alexander Afeyan[a,b], Jasmin Mraz[a,c], Elinor Ng Eaton[a], Ferenc Reinhardt[a], Yevgenia L. Khodor[d], Prathapan Thiru[a], Brian Bierie[a], Xin Ye[a,e], Christopher B. Burge[d], and Robert A. Weinberg[a,f,g,1]

Yu *et al.* Science 2013
Kroger *et al.* PNAS 2019

Hybrid E/M: the ‘fittest’ for metastasis?



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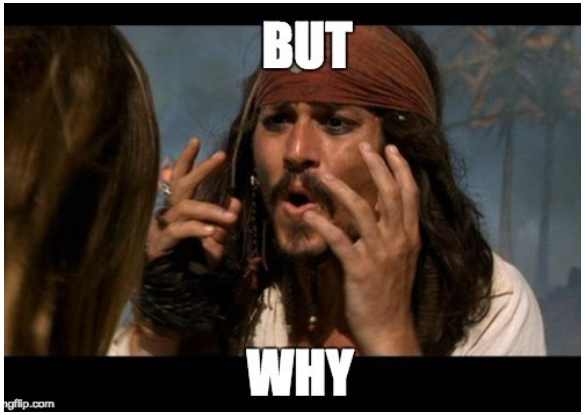
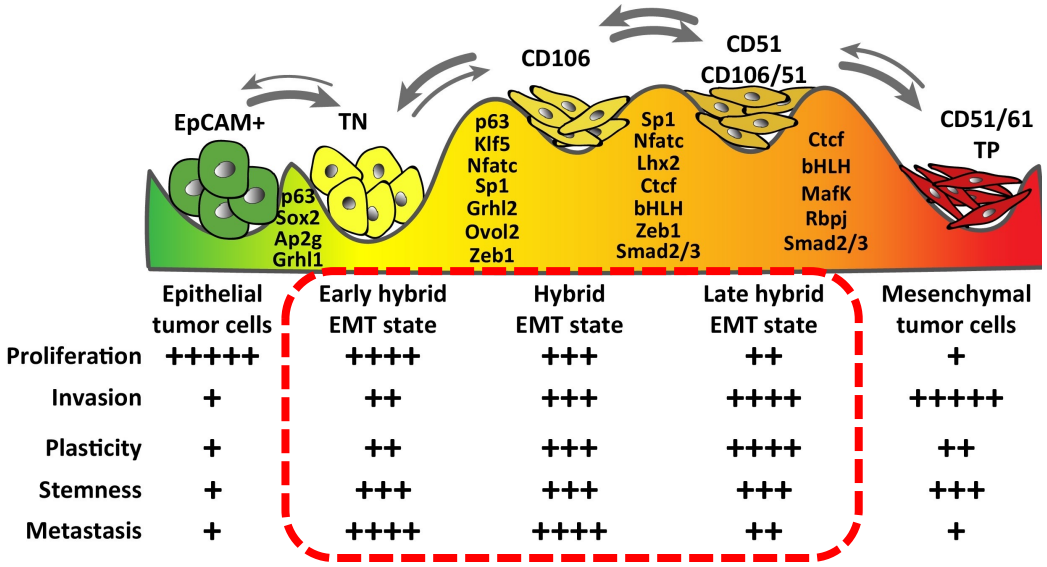


Why are hybrid E/M cells relatively more plastic?

Pastushenko *et al.* 2019

(*Man vs. Wild* TV series – Bear Grylls; Pirates of Carribean)

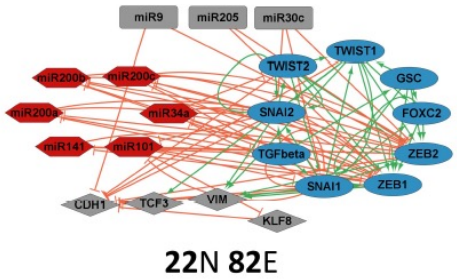
Why are hybrid E/M cells the ‘fittest’?



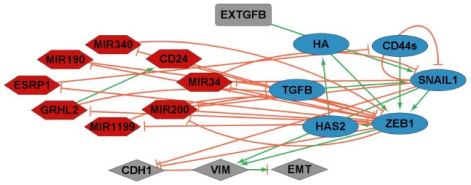
Kishore Hari
(PhD, IISc)

Hari *et al.* eLife 2022

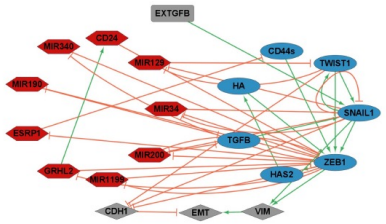
Is the “high plasticity” behavior of hybrid E/M a feature of underlying regulatory networks?



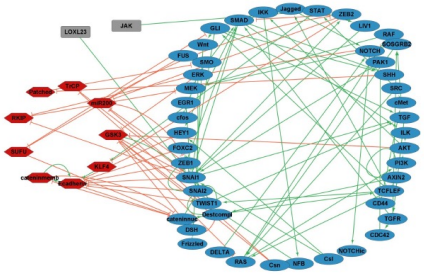
22N 82E



18N 33E



20N 40E



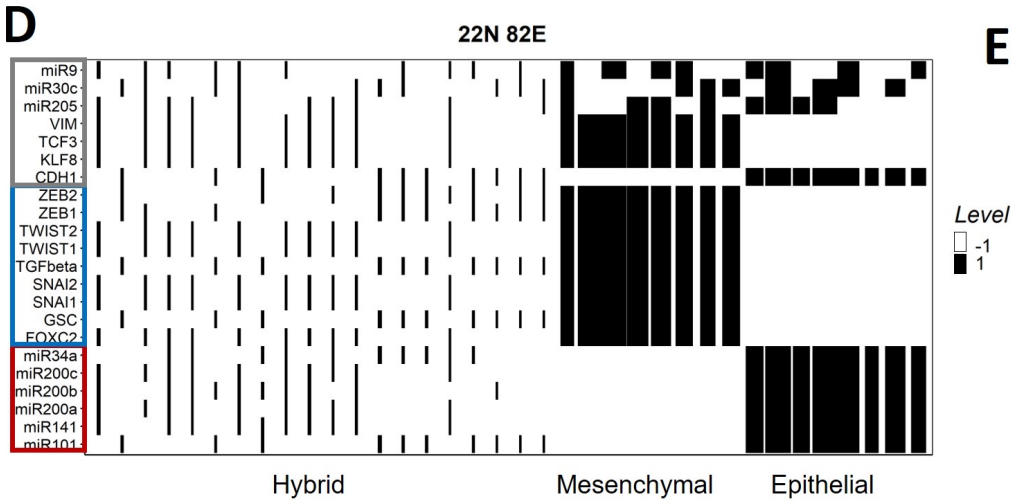
57N 113E

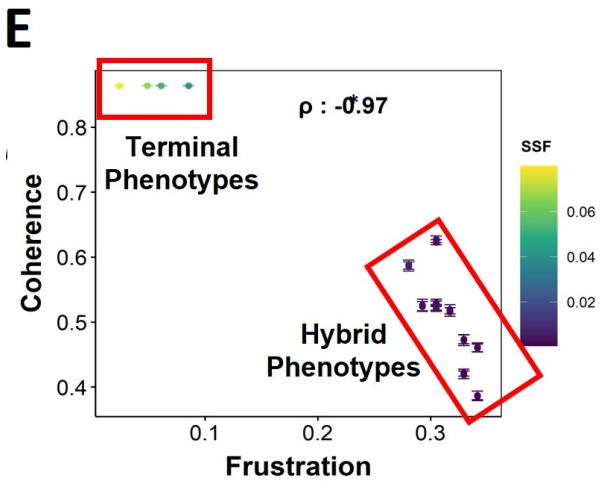
Nodes: Epithelial, Mesenchymal.

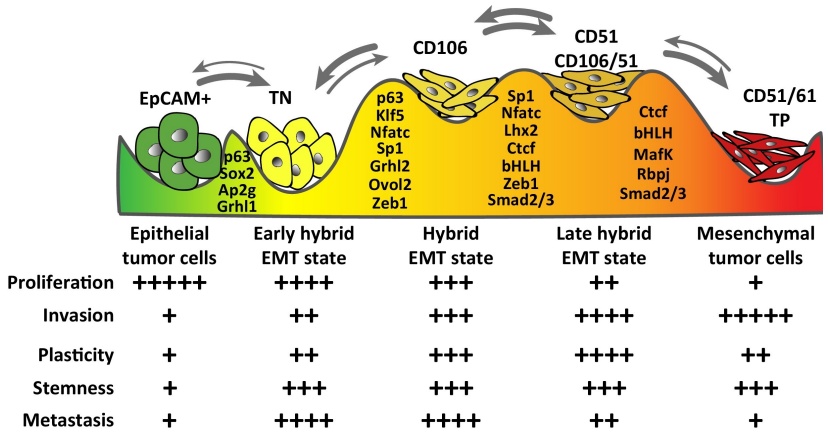
Edges: Activation, Inhibition

EMP networks allow for two sets of states

$$s_i(t+1) = \begin{cases} +1, \sum_j Adj_{ij} s_j(t) > 0 \\ -1, \sum_j Adj_{ij} s_j(t) < 0 \\ s_i(t), \sum_j Adj_{ij} s_j(t) = 0 \end{cases}$$







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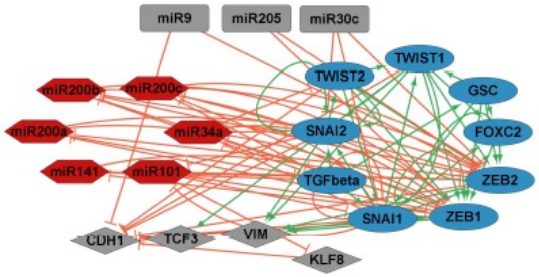
Metastasis	+	++++	++++	++	+

Hybrid E/M states are more frustrated than E, M. But why?

Hari *et al.* eLife 2022

EMT networks consist of two “teams” of players



22N 82E

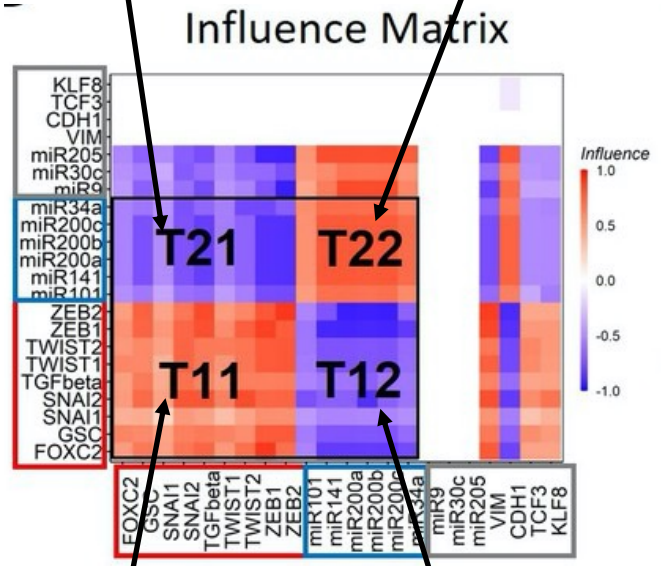
$$Infl = \frac{\sum_{l=1}^{lmax} \frac{Adj^l}{Adj^l_{max}}}{lmax}$$

$$T_{KL} = \frac{\sum_{i \in T_K} \sum_{j \in T_L} Infl_{ij}}{n_{KL}}, K, L \in \{1,2\}$$

$$Ts = \frac{\sum_{K,L \in \{1,2\}} |T_{KL}|}{4}$$

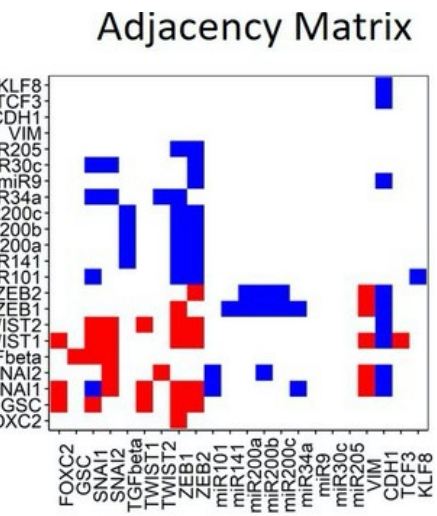
All Epi nodes effectively **inhibit** all Mes nodes.

All Epi nodes effectively **activate** all Epi nodes.



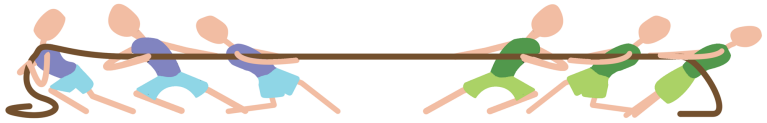
All Mes nodes effectively **activate** all Mes nodes.

All Mes nodes effectively **inhibit** all Epi nodes.

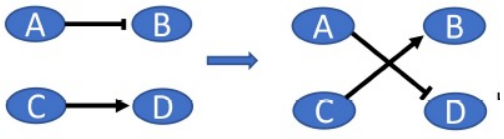


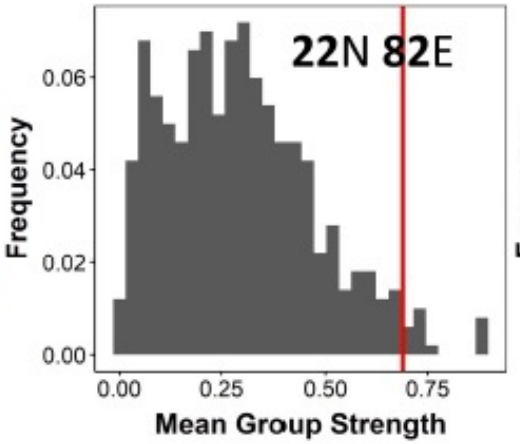
Row => Source; Column => Target

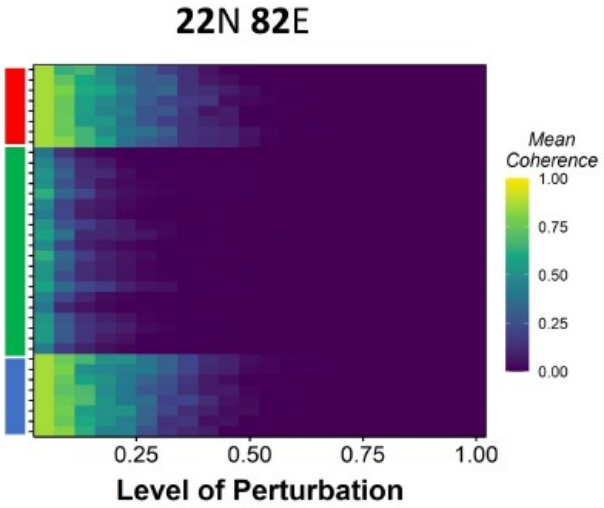
Red (Activation), Blue (Inhibition)

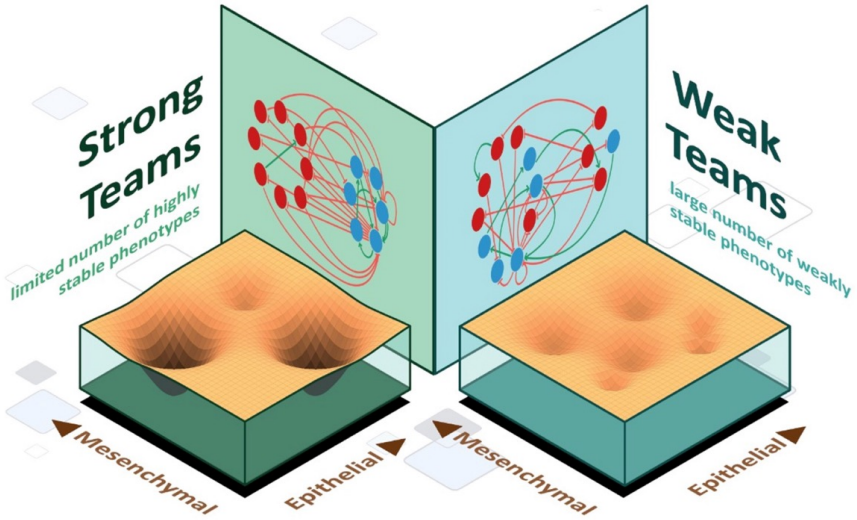


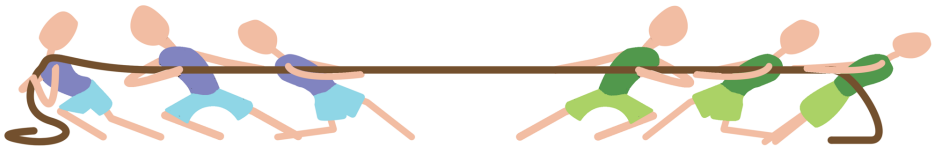
The presence of “teams” is specific to EMP networks











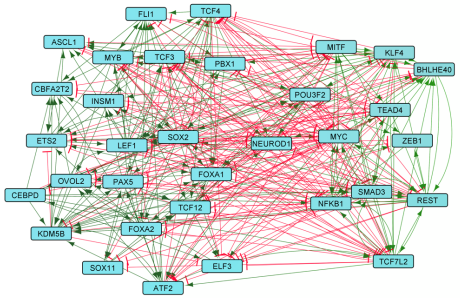
Absence of any “teams” supporting the hybrid E/M phenotypes makes them the ‘fittest’ for metastasis.

Hari *et al.* eLife 2022

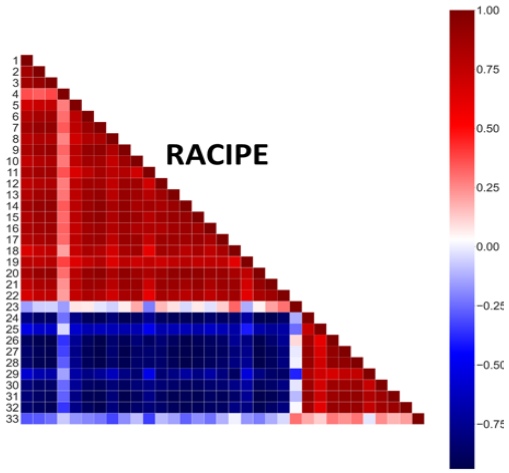
Are “teams” seen in other examples of plasticity too?

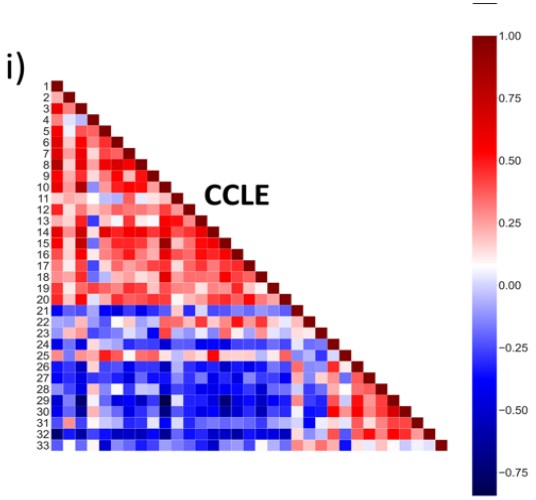
Model prediction

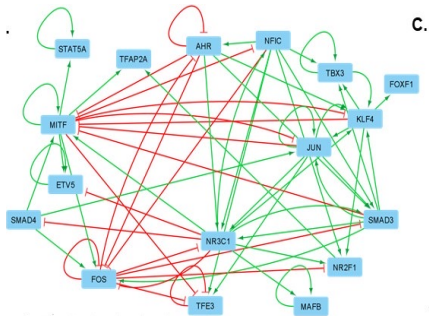
Experimental validation



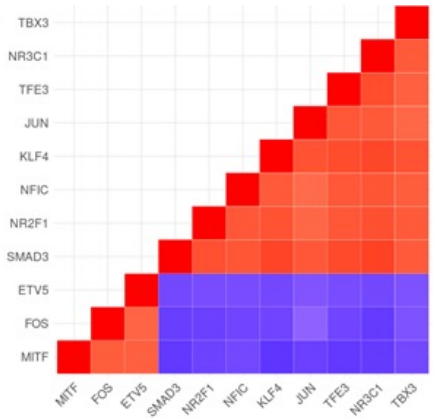
Small Cell Lung Cancer
(33N 357E)

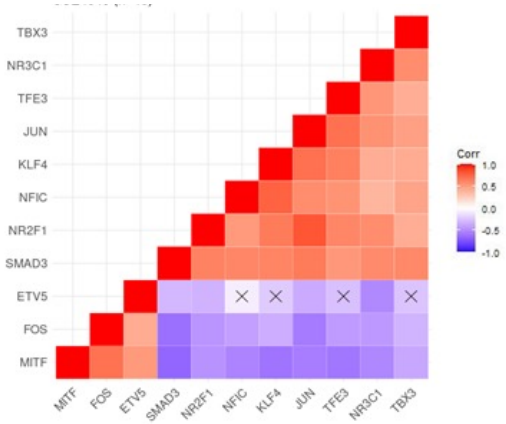






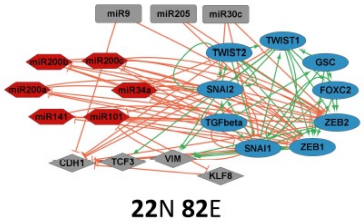
Melanoma (17N 52E)



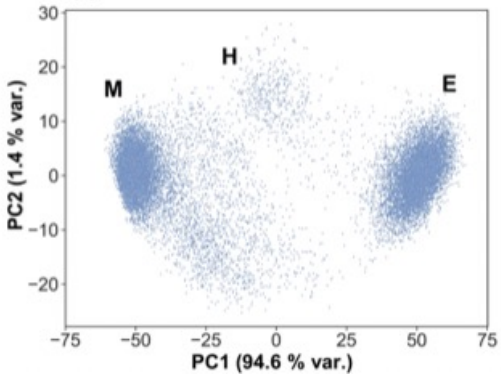


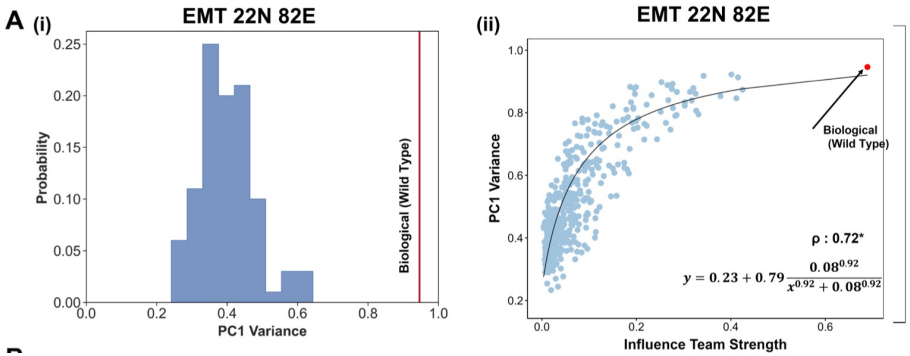
Pillai & Jolly, iScience 2021
Chauhan*, Ram*, Hari & Jolly, eLife 2021

“Teams” – a meaningful dimension-reduction metric?



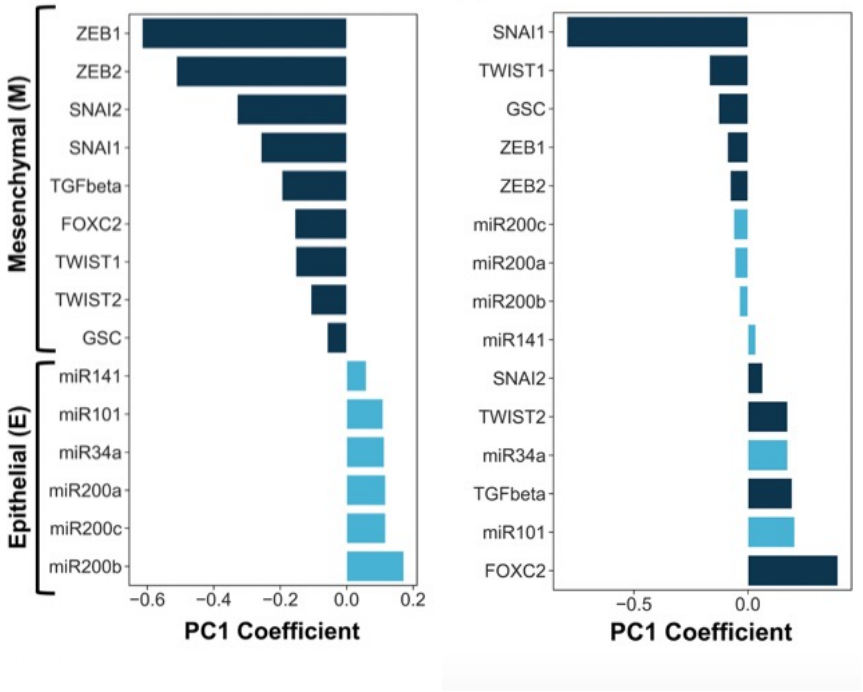
22N 82E





WT network

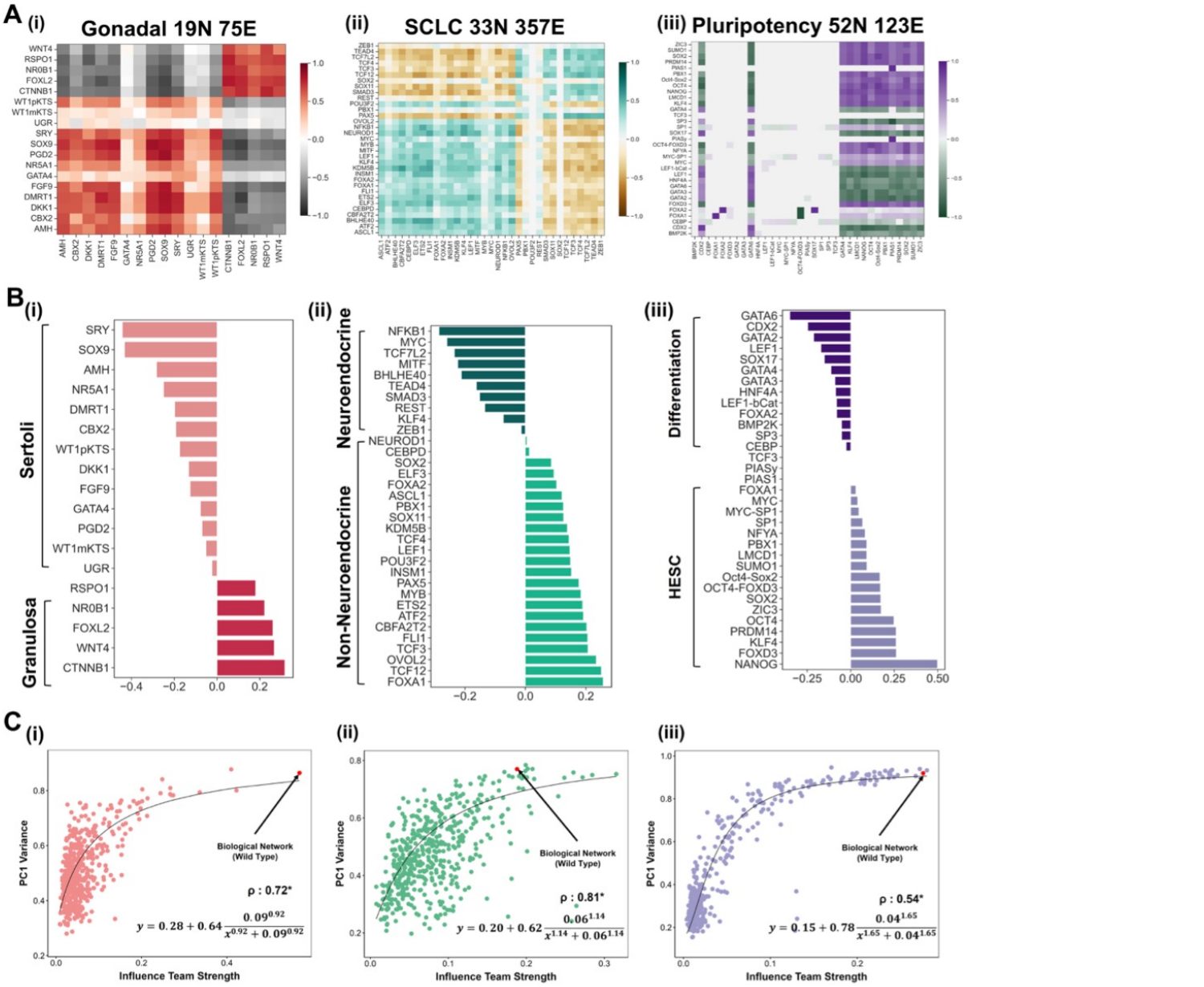
Negative control



- EMT networks or transcriptomic data can be explained mostly by PC1.

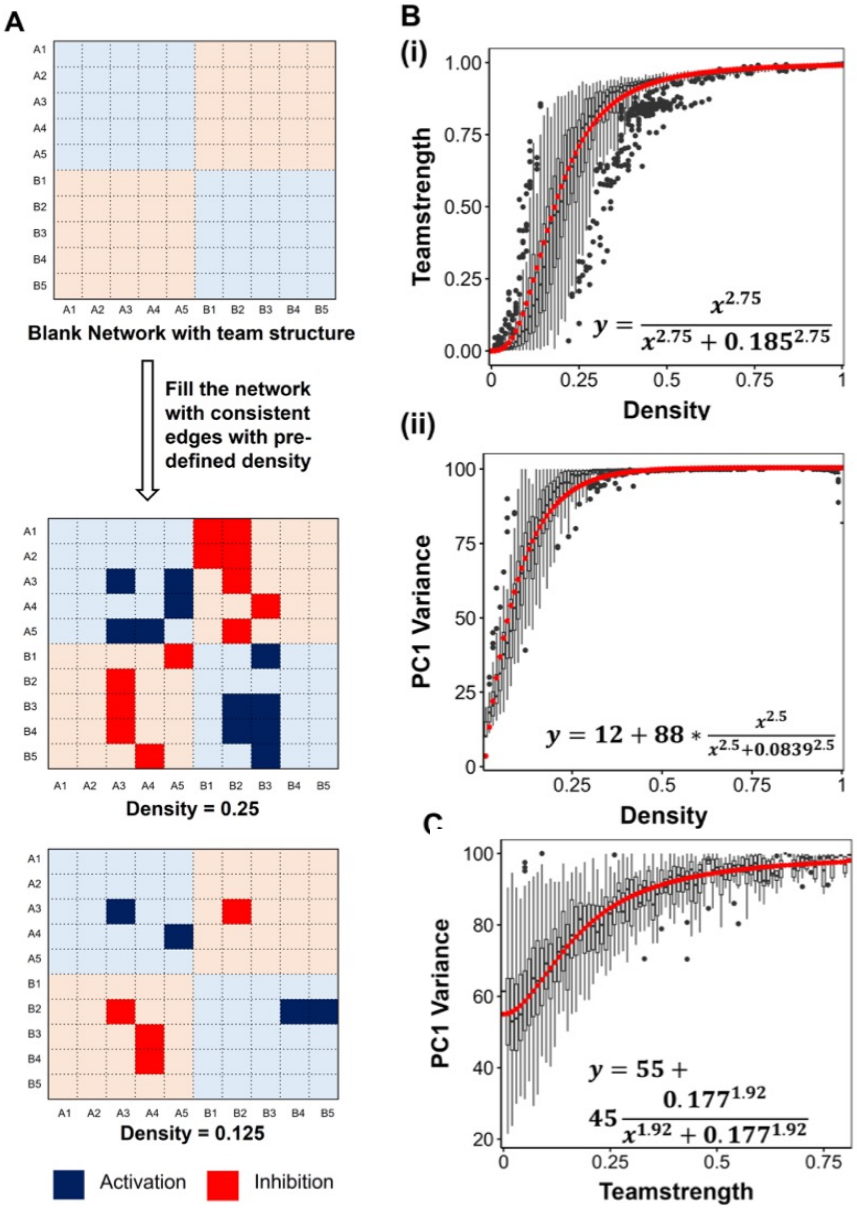
Hari *et al.*, bioRxiv 2023: 526930

Low-dimensionality of phenotypic space : other examples



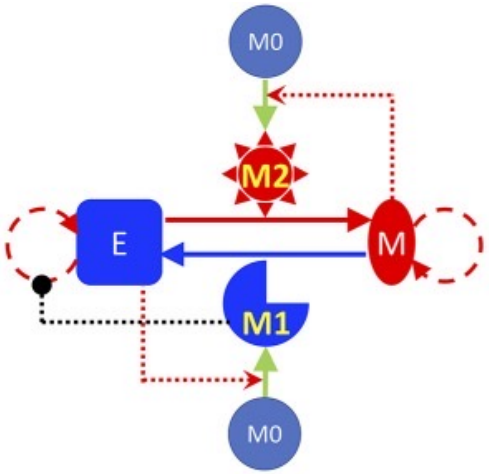
Hari *et al.*, bioRxiv 2023: 526930

Impact of “teams” on canalization can be generalized



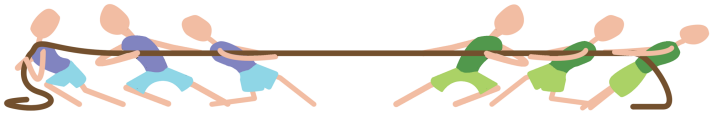
Hari *et al.*, bioRxiv 2023: 526930

From “teams” of nodes in a cell to “teams” of cells in a tissue



Two states of the cell population model:

1. Epithelial cancer cells, M1 macrophages
2. Mesenchymal cancer cells, M2 macrophages



Trends in Cancer

CellPress

Special Issue: Quantitative Cancer Biology

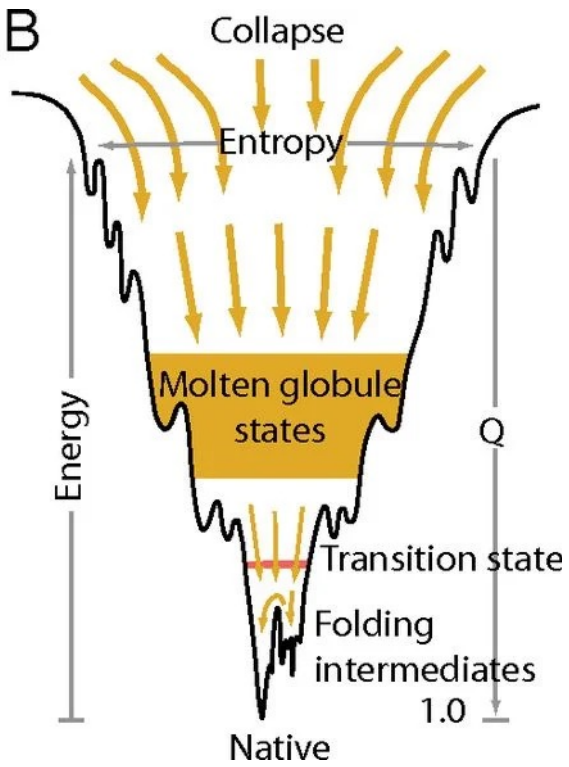
Review

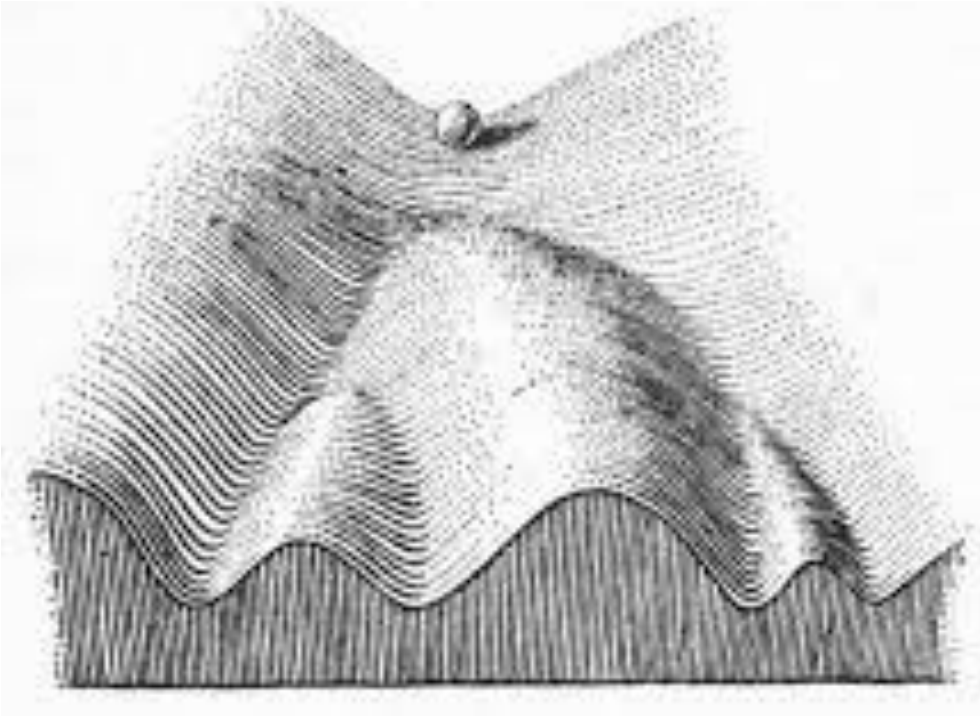
Group Behavior and Emergence of Cancer Drug Resistance

Supriyo Bhattacharya,[1] Atish Mohanty,[2] Srisairam Achuthan,[3] Sourabh Kotnala,[2] Mohit Kumar Jolly,[4] Prakash Kulkarni,[2] and Ravi Salgia[2,*]

Li*, Jolly* *et al.* Front Oncol 2019

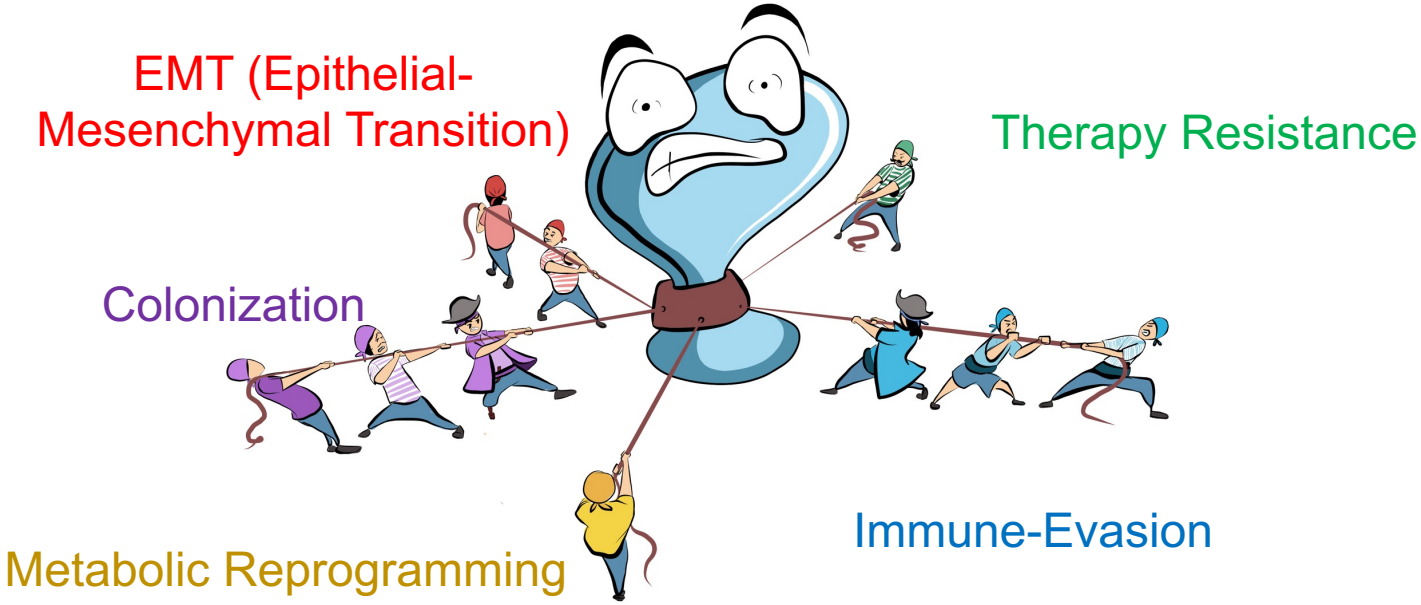
“Teams” ~ driving principle of cell-fate canalization?

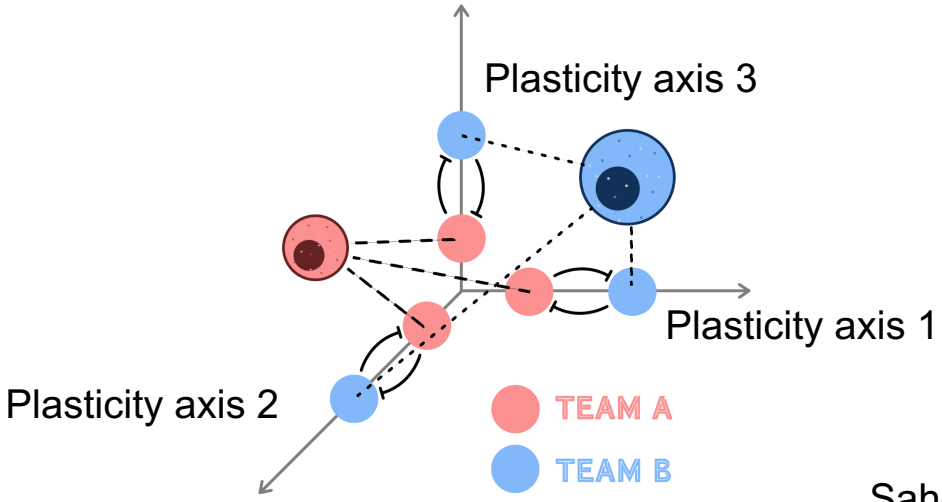




Englander & Mayne, PNAS 2014; Waddington, 1942

Can “teams” help coordinate many axes of plasticity?

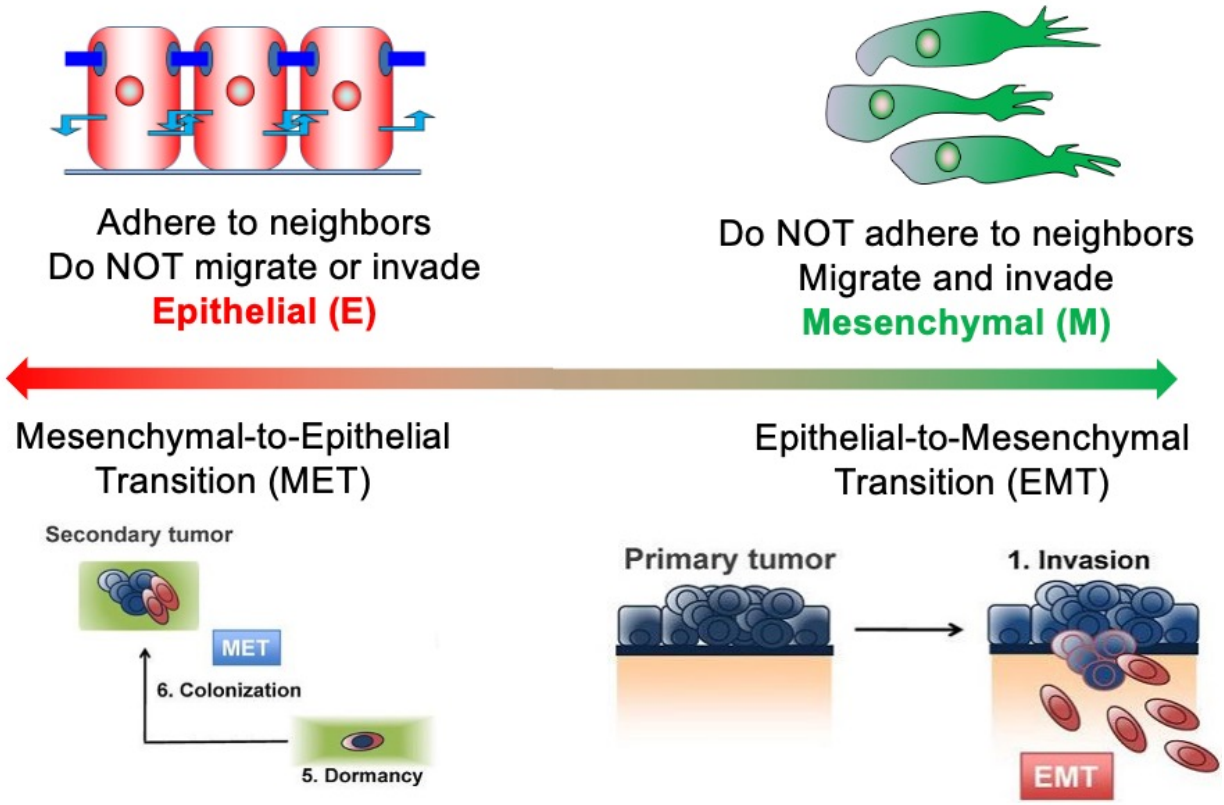




Sahoo *et al.*, manuscript in preparation

Tamoxifen resistance in ER+ breast cancer

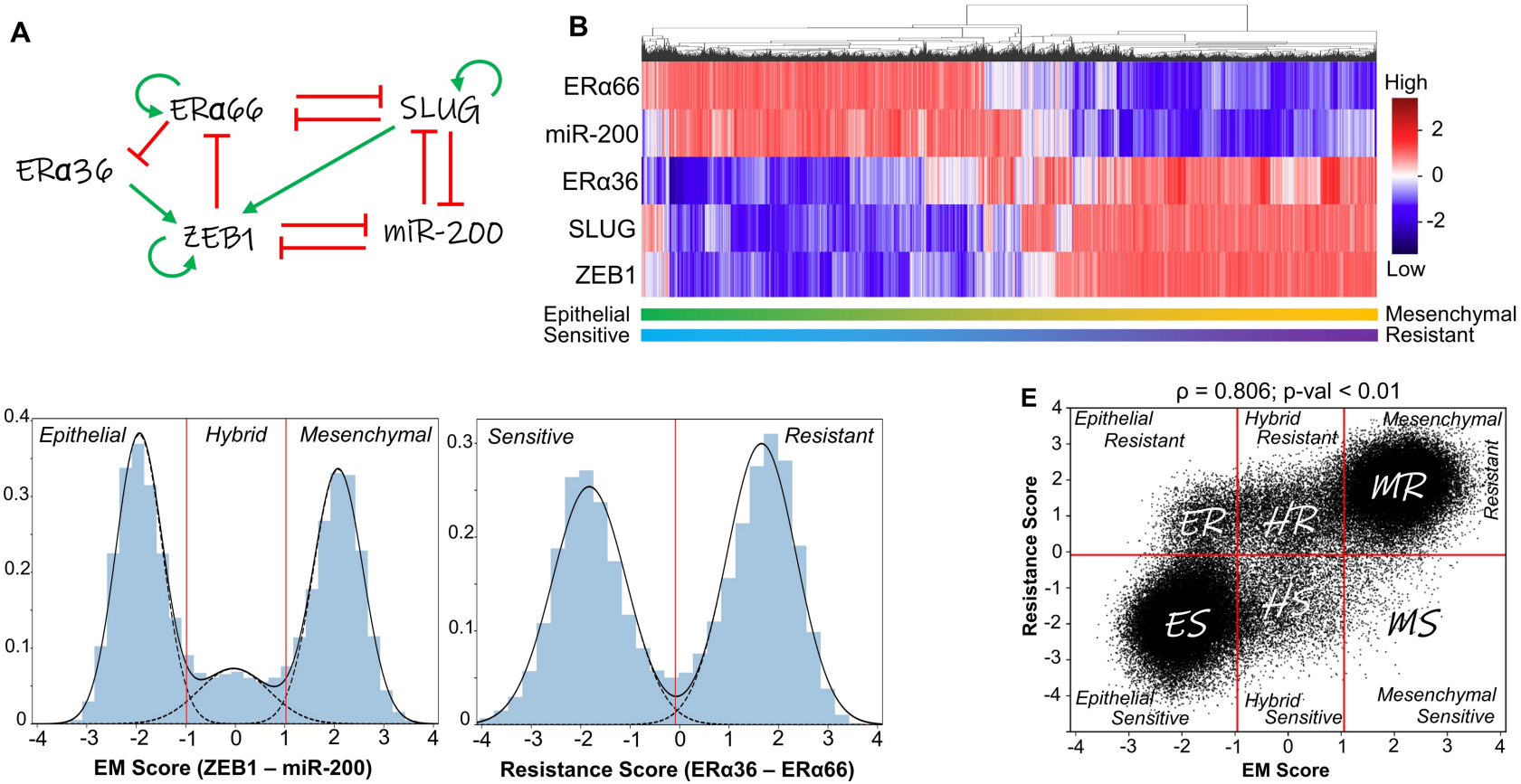
Tamoxifen: 1st targeted therapy; given to ER+ breast cancer patients (75% of BC cases)



- Does EMT drive tamoxifen resistance or *vice versa*?
- Can state-switching enable long-term 'resistance' without genetic changes?

Scheel & Weinberg, Seminars in Cancer Biology 2012

Association between (E, sensitive) and (M, resistant) states

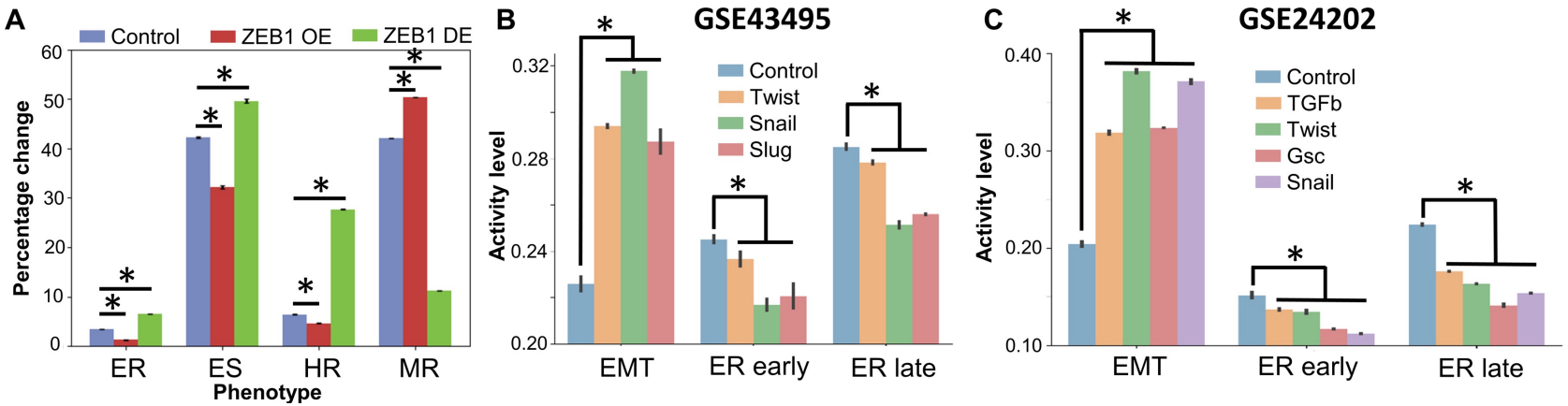


- E state usually Tam-Sensitive; M state usually Tam-Resistant
- Hybrid E/M state can be Tam-Resistant too

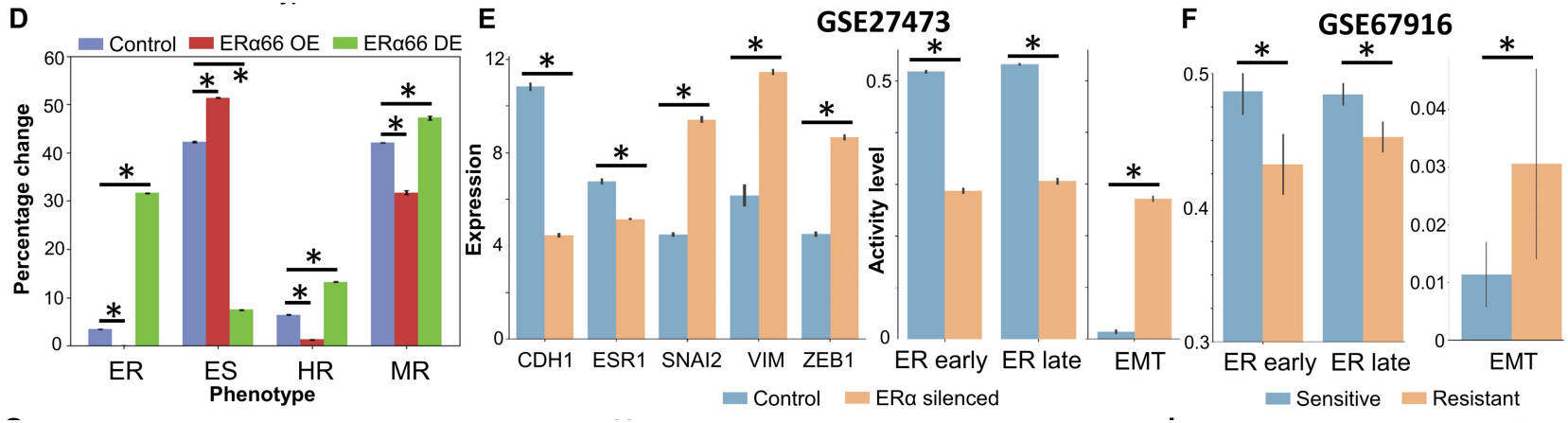
Sarthak Sahoo

Sahoo *et al.* NAR Cancer 2021

EMT & Tam Res can drive each other



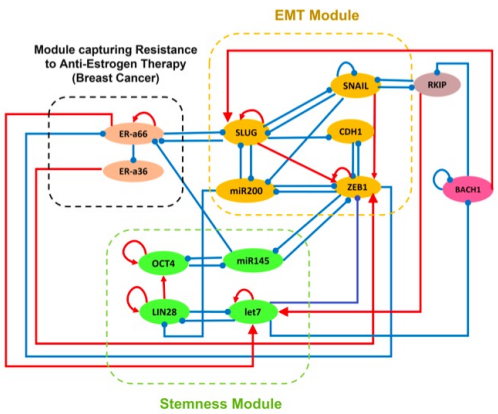
EMT can drive resistance to tamoxifen

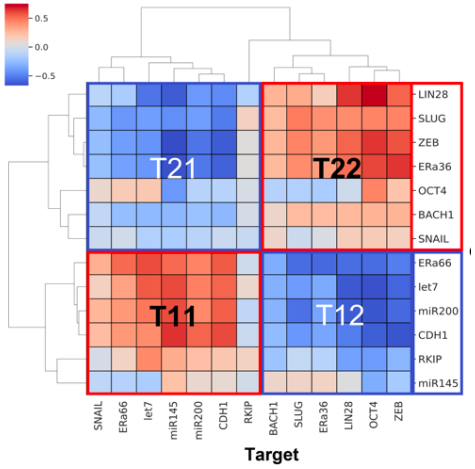


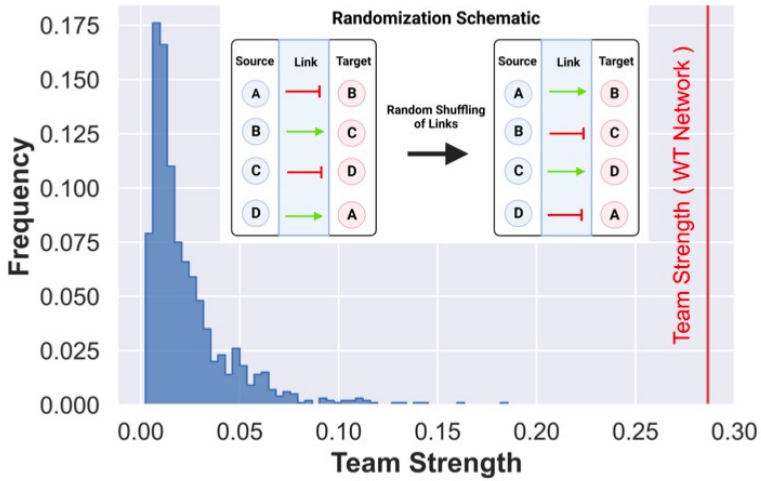
Tamoxifen resistance can drive EMT

Sahoo *et al.* NAR Cancer 2021

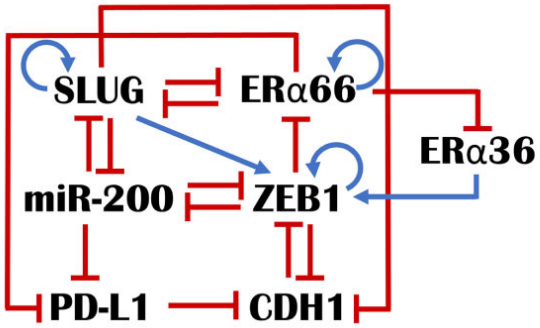
“Teams” enabling coupling between more than 2 axes

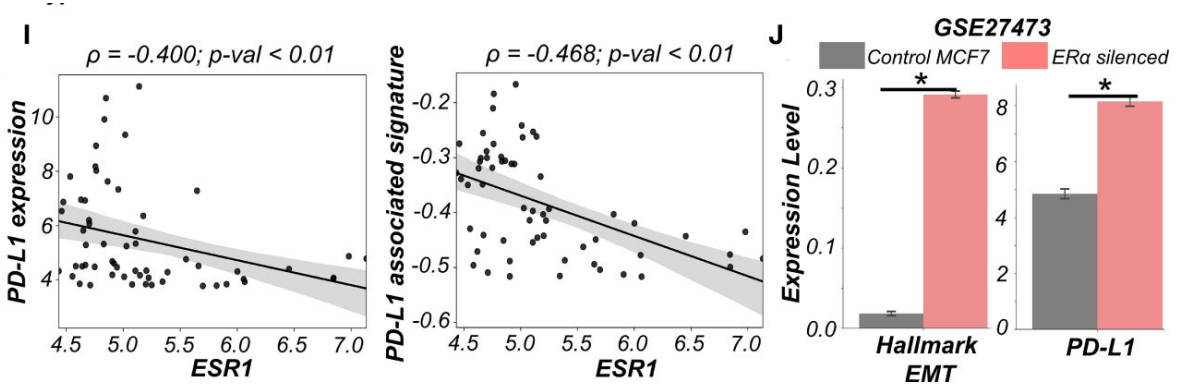






“Teams” connecting EMT, Tam-Res and stemness phenotypes

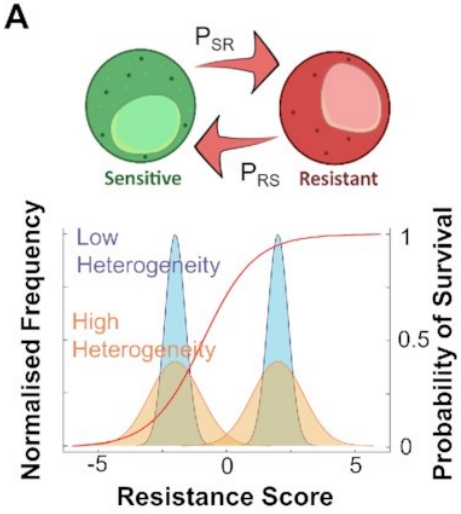


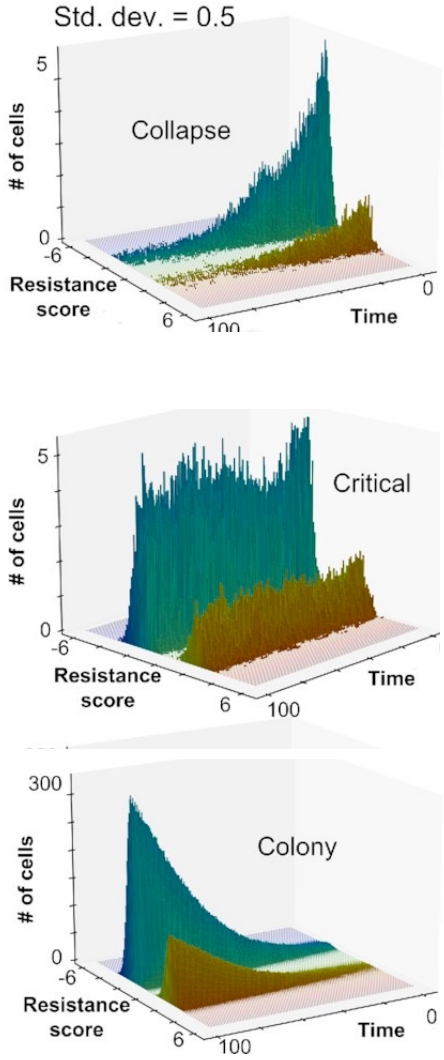


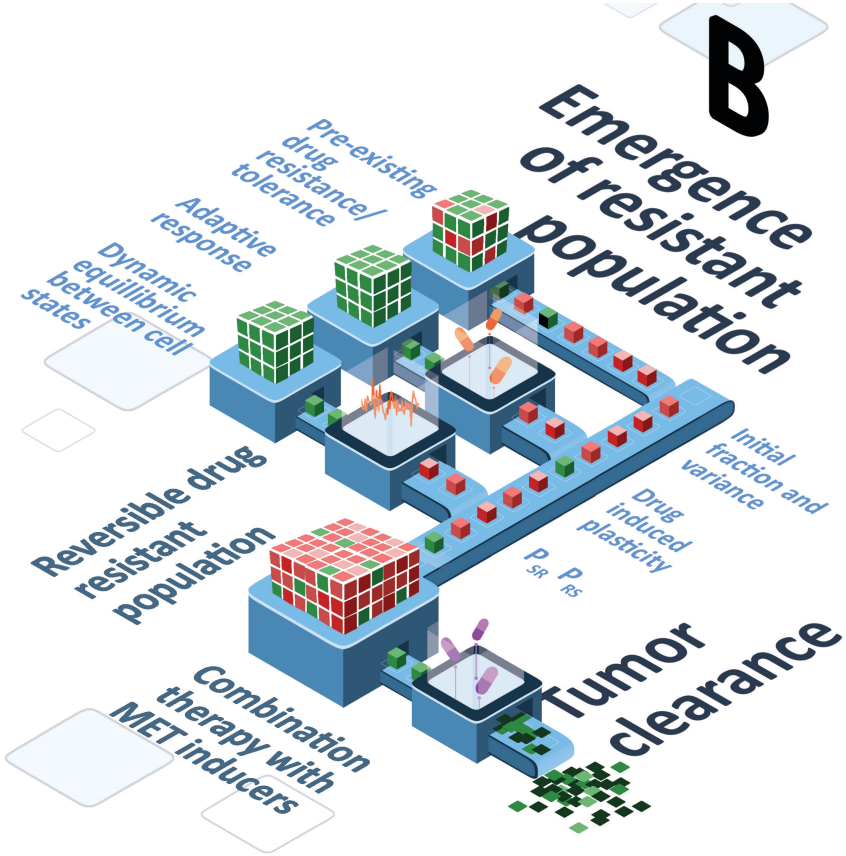
“Teams” connecting EMT, Tam-Res and PD-L1 (+ve) phenotypes

Sahoo *et al.* Front Immunol 2021; Shyam *et al.* J R Soc Interface 2023

Suggesting combinatorial therapies for ER+ breast cancer





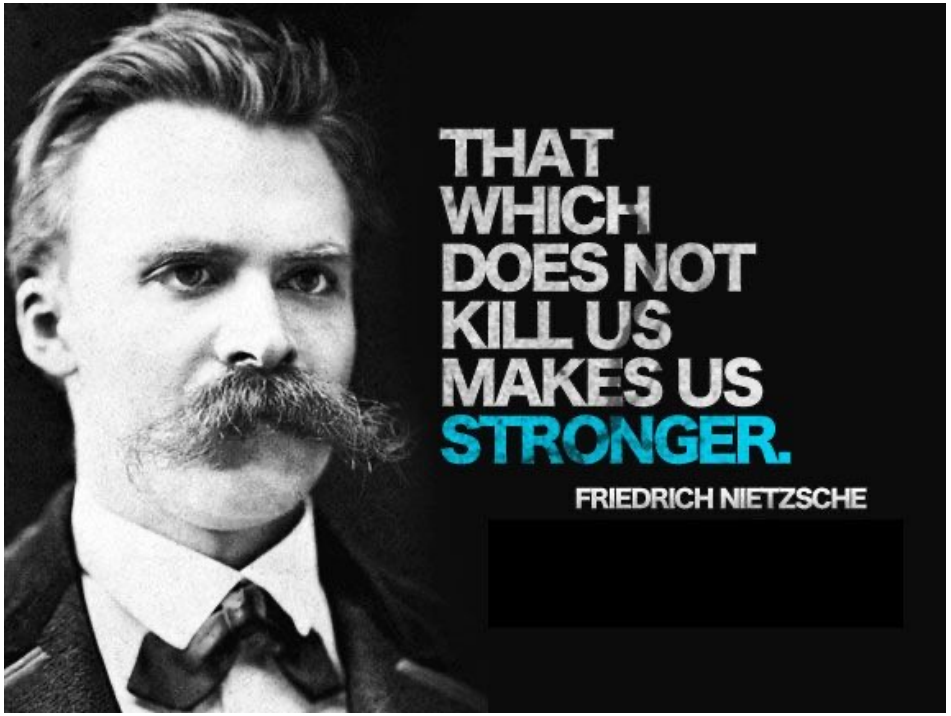


Model predictions currently undergoing experimental validation

Sahoo *et al.* NAR Cancer 2021

A population of cancer cells exposed to a targeted therapy/ immunotherapy can:

a) Die
b) Become dormant transiently
c) Switch to a more aggressive behavior
d) …

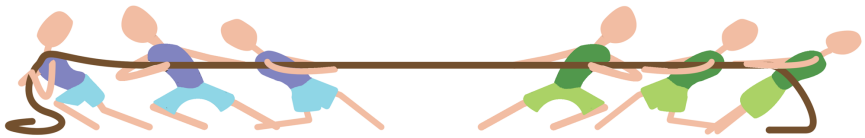


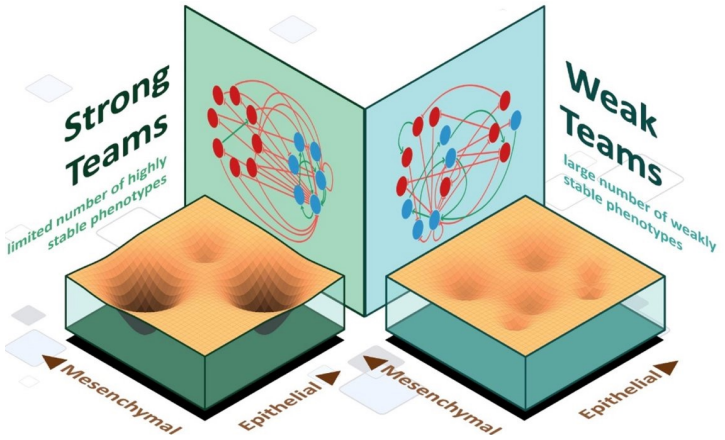
Avada Kedavra
(Targeted therapy)

Cells that adapt & survive:
Hail the “Team” Potter!

Summary (Part 2)

- Regulatory networks underlying cancer cell plasticity are multi-stable.
- “Teams” control relative stability (or lack thereof) of diverse phenotypes.
- “Teams” can coordinate various axes of plasticity (EMT, TamRes *et al.*)
- Such coordination can facilitate therapy-driven adaptive responses, aggravating clinical outcomes.

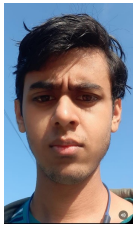




Open questions

What if there are more than two “teams” in action?

Vaibhav Anand (IISER-P)



Kushal Haldar (IISER-K)

Aditya Moger (IISER-P)

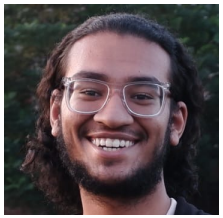
(Game of Thrones)

Open questions

Do “teams” offer more robustness against fluctuations?

Lakshmi Malvadi (IISc)

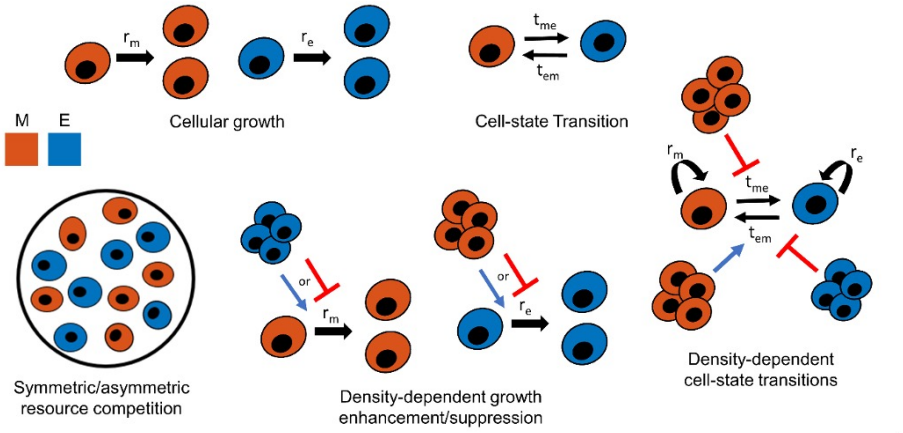
Kishore Hari (IISc; now at Northeastern)

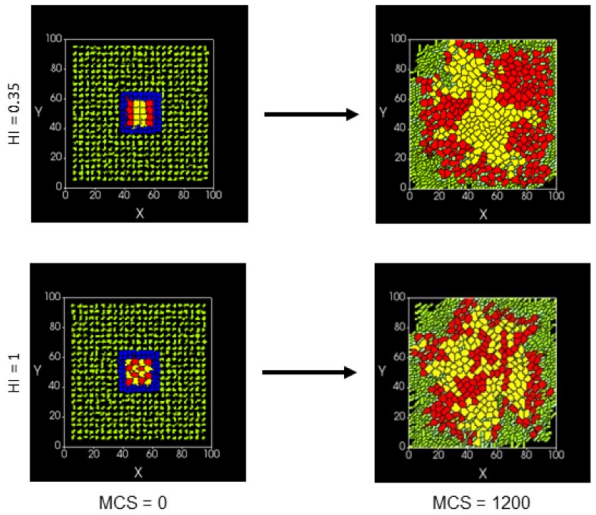


Abhay Gupta (IISER-M)

(Mr. Bean)

Ongoing work





How do rates of cell-state transition depend on prior history & neighborhood?

How does spatial heterogeneity impact cancer invasion traits?

Paras Jain
(PhD, IISc)

Collaborators:
Jason A George (TAMU)
Rik Thompson (QUT)
Michael Toneff (Widener)

Jain *et al.* Biomolecules 2022
Jain *et al*. J R Soc Interface 2023
Jain *et al.* bioRxiv 2023

CVS Prasanna
(PhD, IISc)

Collaborators:
Ramray Bhat (IISc)
Federico Bocci (UCI)

Pramanik *et al.*
J Theor Biol 2020
Prasanna *et al.*
Biophys J 2024, in press

Acknowledgements: Our "team"!

- Biotechnology
- Electrical Engineering
- Bioinformatics
- Physics
- Mathematics
- Cancer Biology

- Oncologists
- Clinicians
- Mathematicians
- Physicists
- Chemists
- Engineers

ACTREC · ncbs national centre for biological sciences · tifr TIFR Centre for Interdisciplinary Sciences

St. John's Research Institute · City of Hope · Duke University · A&M

Montana State University · Queensland University of Technology · Northeastern University · Northwestern University

ICR The Institute of Cancer Research · Garvan Institute of Medical Research

Widener University · IIT Bombay · icmr NIRRCH

Funding: SERB India · MHRD · Infosys Foundation · Wellcome Trust

To join our team; contact: mkjolly@iisc.ac.in