Theory

Cell

Stochastic State Transitions Give Rise to Phenotypic Equilibrium in Populations of Cancer Cells

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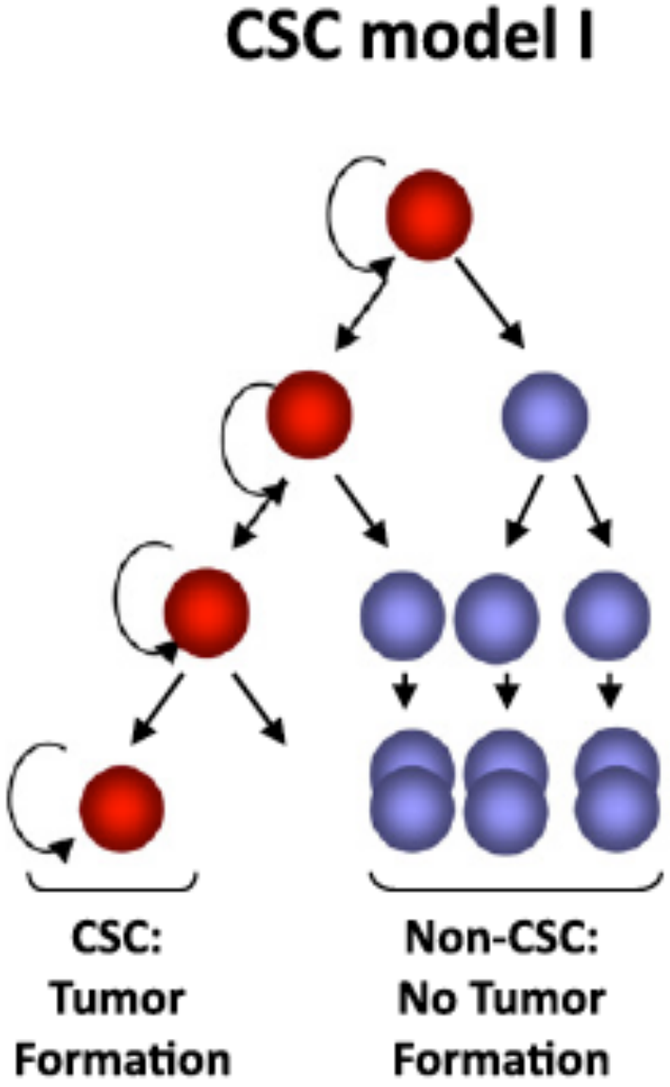
Cell *146*, 633–644, August 19, 2011

Cancer cells within individual tumors:

- distinct phenotypic states, different functional attributes
- equilibria in proportions of cells in the different states

➡ mechanism largely unknown

The classical picture



Cancer stem cells (CSC)

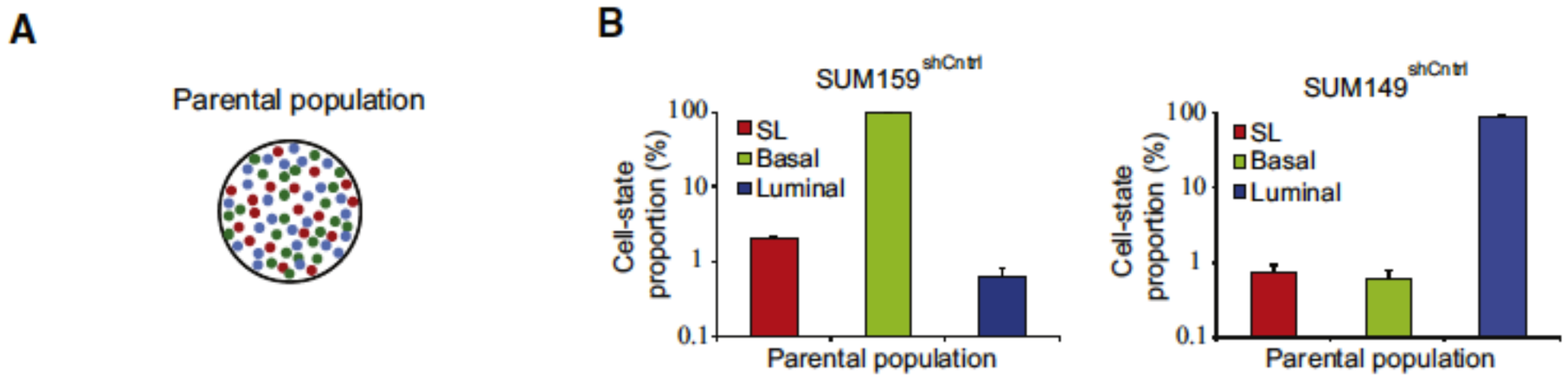
- one of phenotypical states
- give rise to non-CSCs during tumor growth
- non-CSCs cannot produce CSCs
- CSC differentiation gives rise to cell-state equilibrium
- only CSC seed tumors

▸ hierarchical cell-lineage structure like in normal tissue development

The experiment

Two breast-cancer cell lines

- three cell phenotypes: basal, luminal and stem-like cells
- exhibit distinct cell-state equilibria



- 99% purification by FACS (fluorescence-activated cell sorting)
- 6 days of cell growth (ca. 6 cell cycles)

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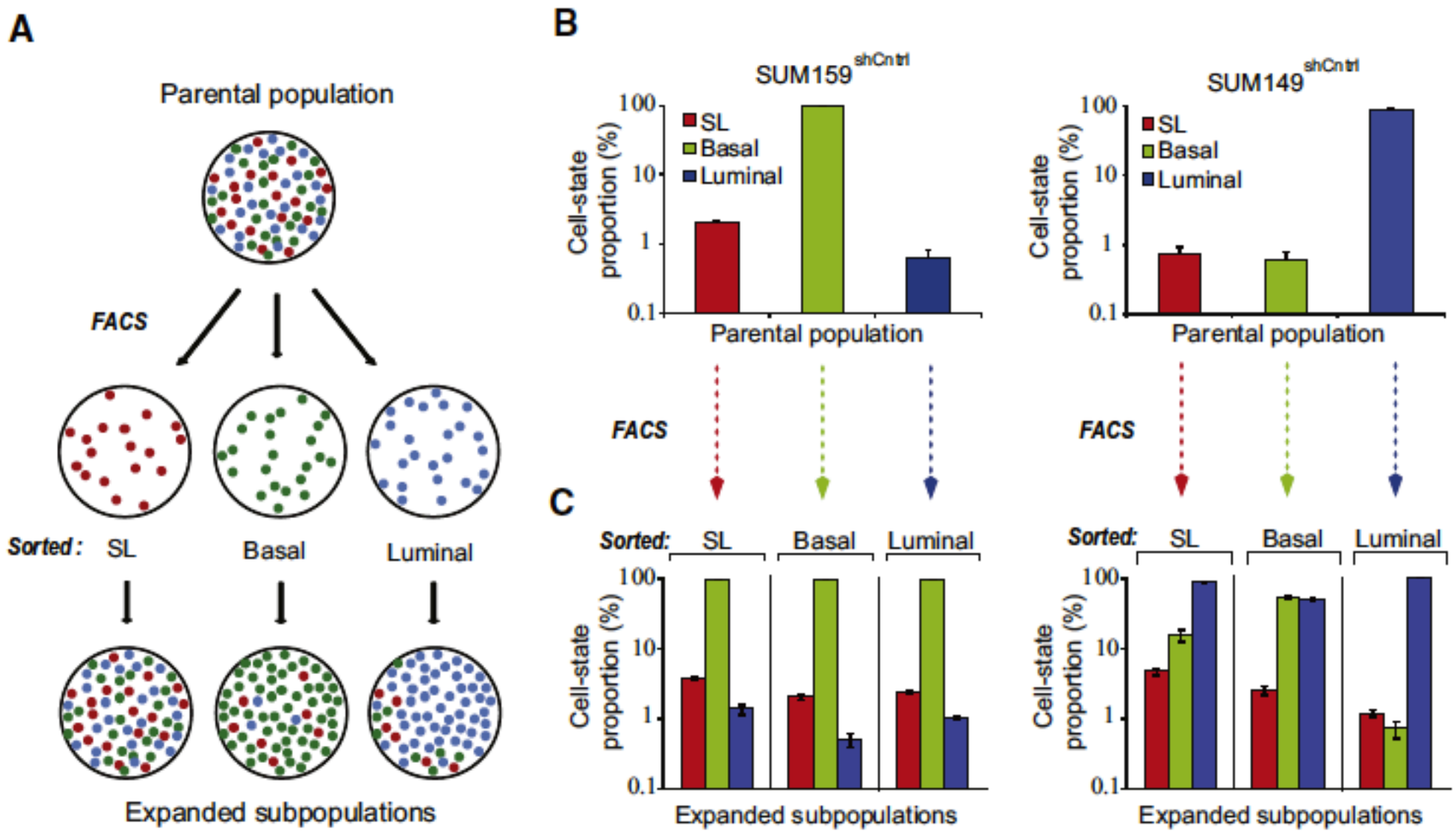

- approach equilibrium in few days (SUM159 faster than SUM149)

Possible explanations?

Frequency-dependent rate of cell growth

- no interconversion of cell states
- cell signaling to detect phenotypic composition of culture

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Frequency-dependent rate of cell growth

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- cell signaling to detect phenotypic composition of culture
- BUT:

  no significant difference in proliferation rate of cell populations (~1 doubling / day)

  "taking over" of population by rare subtype (after FACS) would require doubling rates of 2-3/day

Possible explanations?

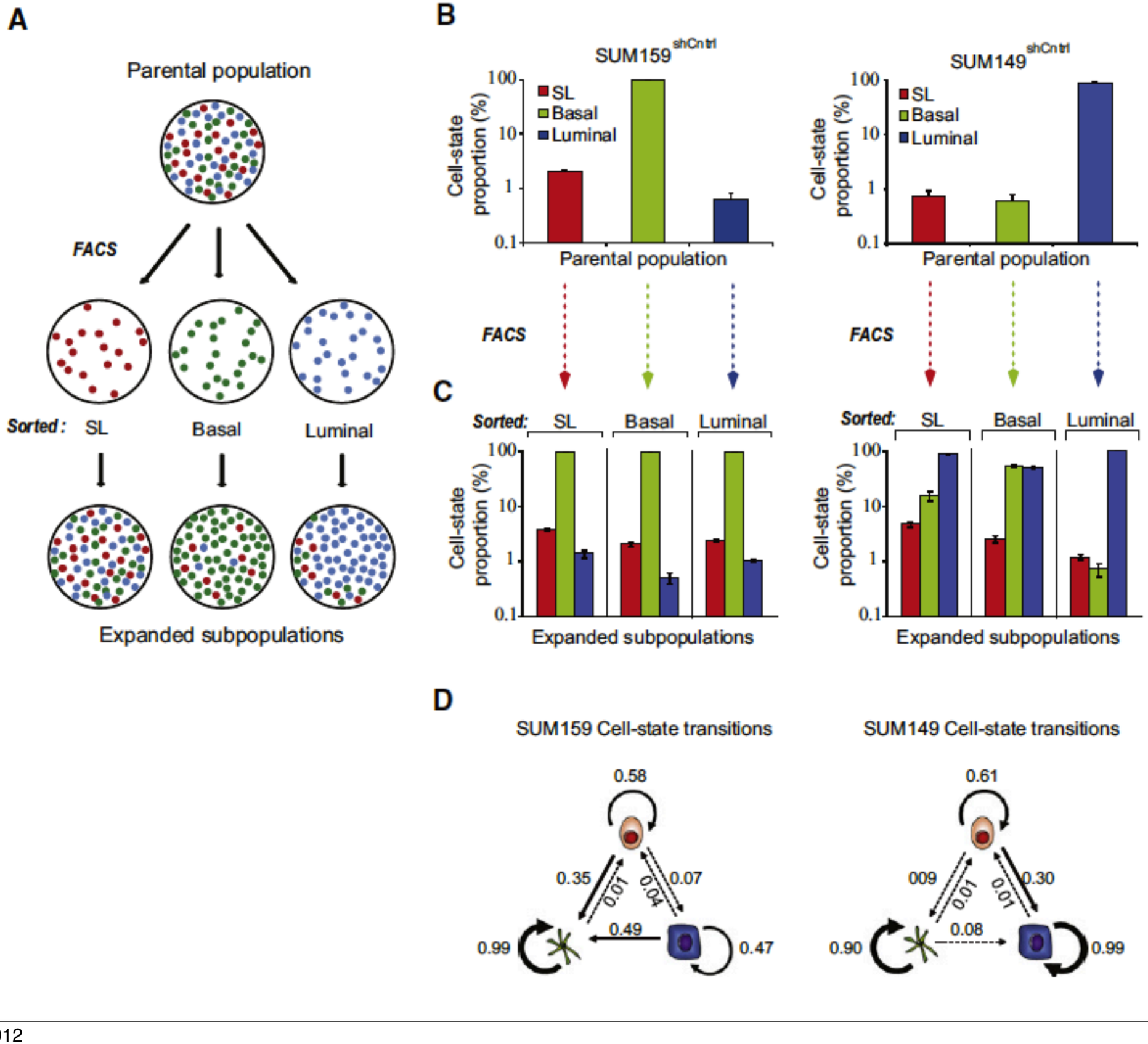
Frequency-dependent rate of cell growth

- no interconversion of cell states
- cell signaling to detect phenotypic composition of culture
- NO!

Interconversion between cell phenotypes

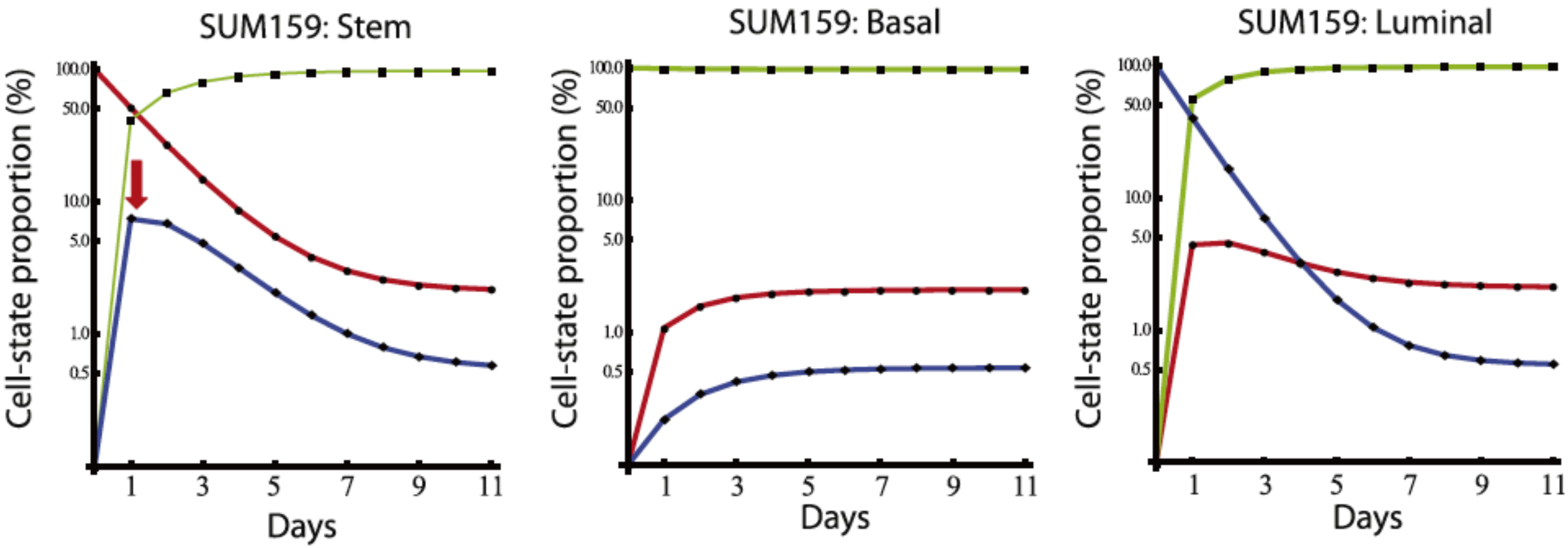
- no interconversion of cell states
- no need for inter-cell communication
- modeling by Markov chains

Possible explanations?



Testing predictions

Markov chain predicts trajectories for phenotypical composition



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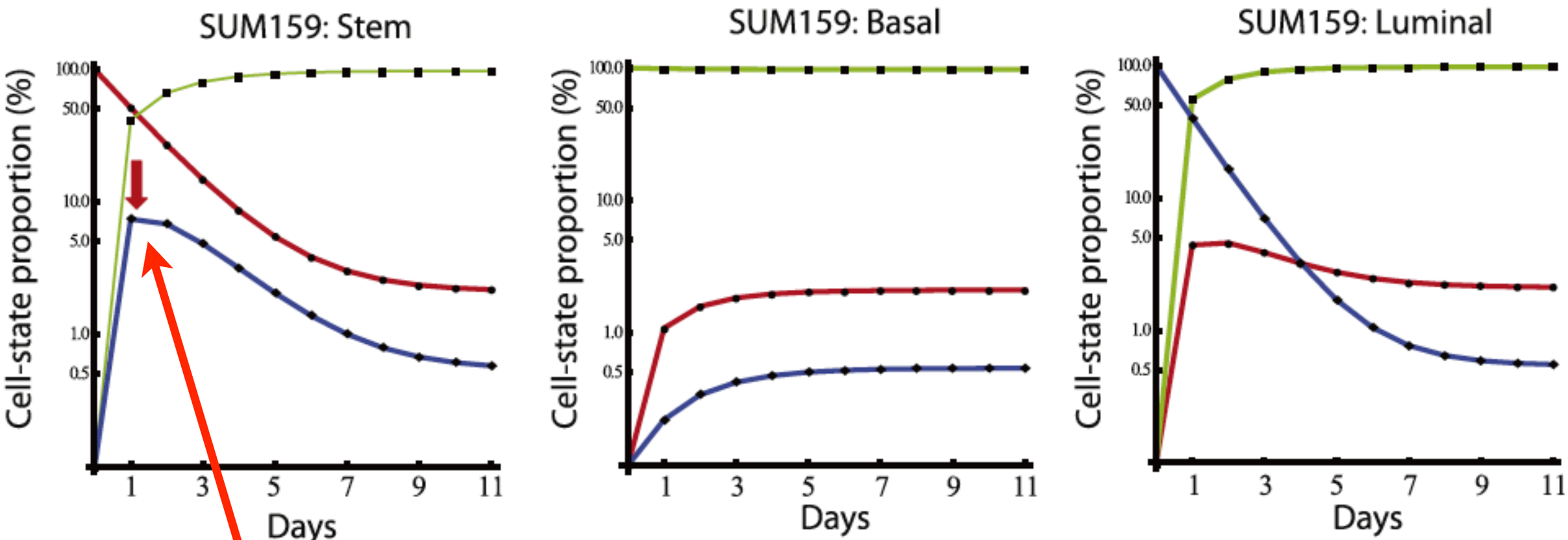

Luminal cells: <1% at day 0 and day 6
transient increase > 7%
Experiment: measured 6.5% at day 1, and 1% at day 6

Tumor seeding capacities

Injection of purified cells into mice:

only stem cells able to induce tumor growth

phenotypical fractions in tumor close to cell line SUM159

Table 2. Incidence and Phenotype Analyses of Tumors Arising from Sorted SUM159 Subpopulations

SUM159 Subpopulations				Analysis of Formed Tumors				
Basal	Stem-like	Luminal	Tumor Incidence	Viable cells (%)	GFP-neg H2K-neg (%)	Basal (%)	Stem-like (%)	Luminal (%)
Direct Injection								
+	–	–	0/4					
–	+	–	4/4	17.11	49.34	93.38	6.03	0.59
–	–	+	0/4					

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With GFP + Irrad. SUM159								
–	–	–	0/4					
+	–	–	4/5	58.1 ± 2.1	56.0 ± 6.0	81.7 ± 5.9	11.4 ± 3.5	6.9 ± 2.4
–	+	–	4/5	53.4 ± 4.7	56.7 ± 7.7	67.2 ± 11.4	24.1 ± 8.0	8.6 ± 3.5
–	–	+	4/5	65.7 ± 6.3	57.7 ± 7.2	82.4 ± 8.1	12.0 ± 6.5	5.6 ± 2.7

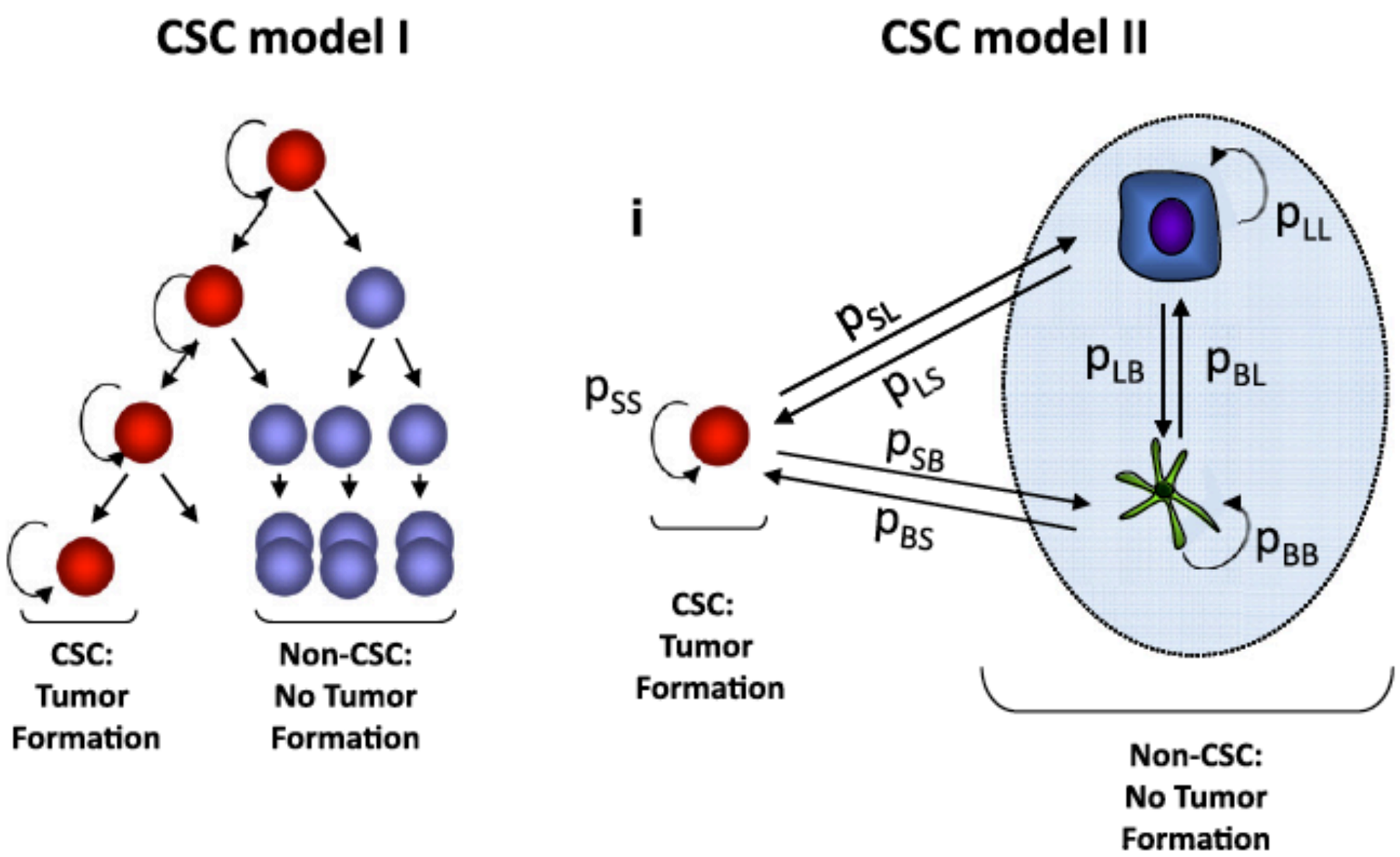
Use irradiated carrier cells:

longer life time

chance to generate new stem-like cells

all three populations able to seed tumor!

A modified picture



Important therapeutic implications!