HERITABLE TUMOR CELL DIVISION RATE HETEROGENEITY INDUCES CLONAL DOMINANCE

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DDS2 Meeting
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INTRATUMORAL HETEROGENEITY

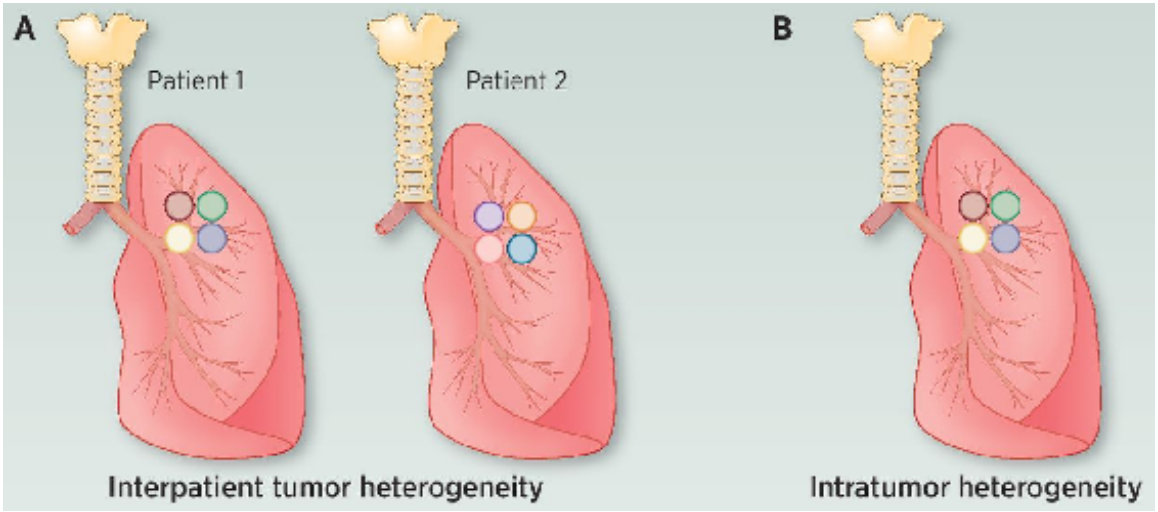


Image from Jamal-Hanjani *et al.*, Clin. Canc. Res., 2015

- Cells within the same tumor vary:
  - variation in environment may cause difference in phenotype
  - variation in genotype/epigenetics may cause difference in phenotype
- Different clones respond differently to treatment

SOURCE OF INTRATUMORAL HETEROGENEITY

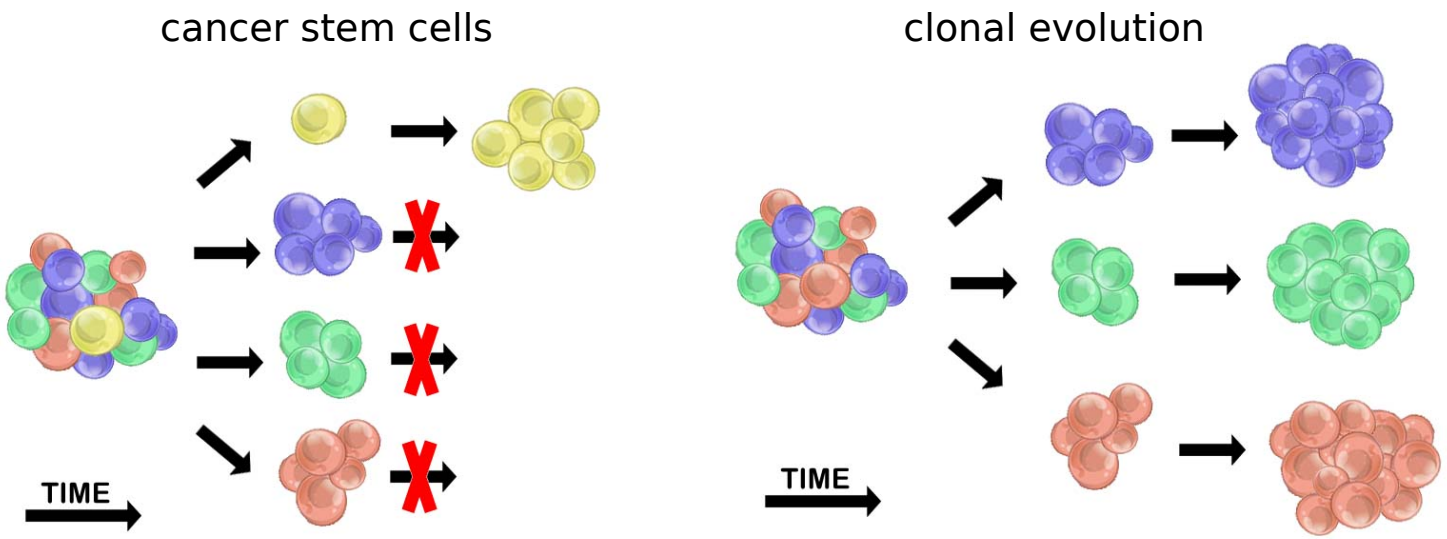
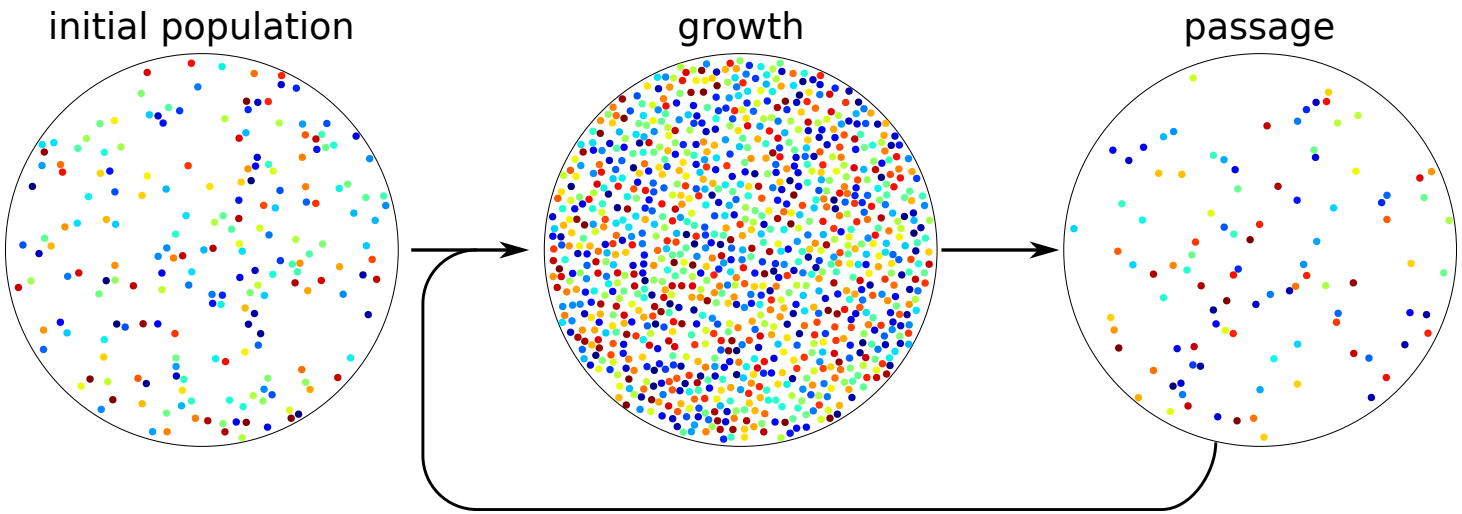


Image adapted from https://en.wikipedia.org/wiki/Tumour_heterogeneity

How to identify the *correct* hypothesis?

- Build computational model for each hypothesis
- Compare:
  - *in vitro* development of tumor cell population (published data)
  - *in silico* development of tumor cell population in matching experiment

ITERATED GROWTH & PASSAGE EXPERIMENT

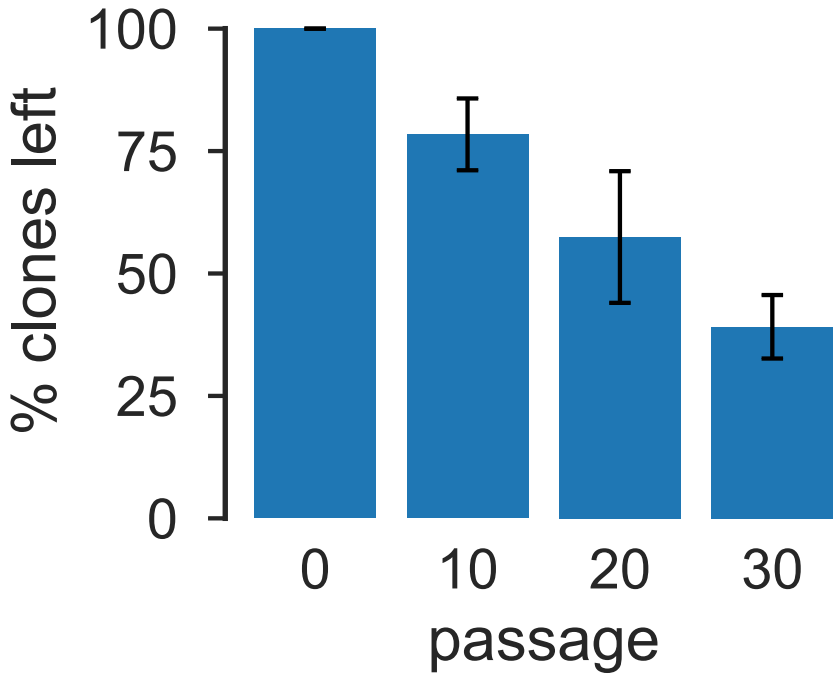


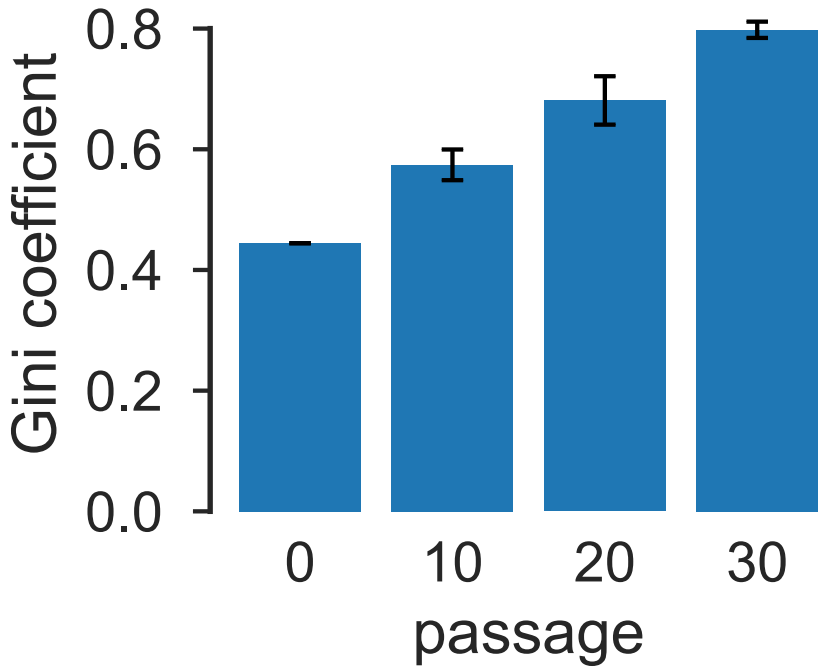
Experimental setup as described in Porter *et al.* (Gen. Biol., 2014):

- Preparation
  - Barcodes are inserted in the DNA using a lentiviral vector
  - Infected cells are selected (using a GFP tag) and grown
  - Three cell populations, each containing $3 \cdot 10^5$, cells are taken
- Experiment (three biological replicates)
  - Each population grows for 3 days, after which $4 \cdot 10^6$ are present
  - $3 \cdot 10^5$ cells are passed on to the next generation

ITERATED GROWTH & PASSAGE EXPERIMENT

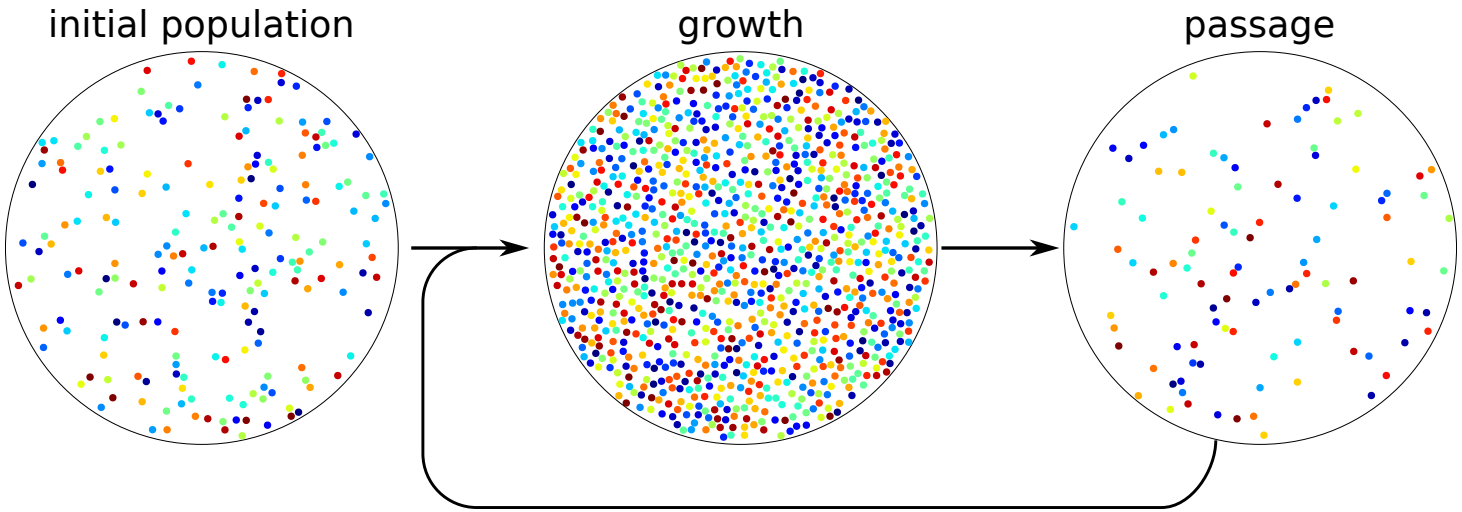
We re-analyzed the experiments from Porter *et al.* (Gen. Biol., 2014), using the FASTQ files in the NIH Sequence Read Archive.





- K562 cell (chronic myelogenous leukemia cell line)
- Clones disappear and clonal dominance increases

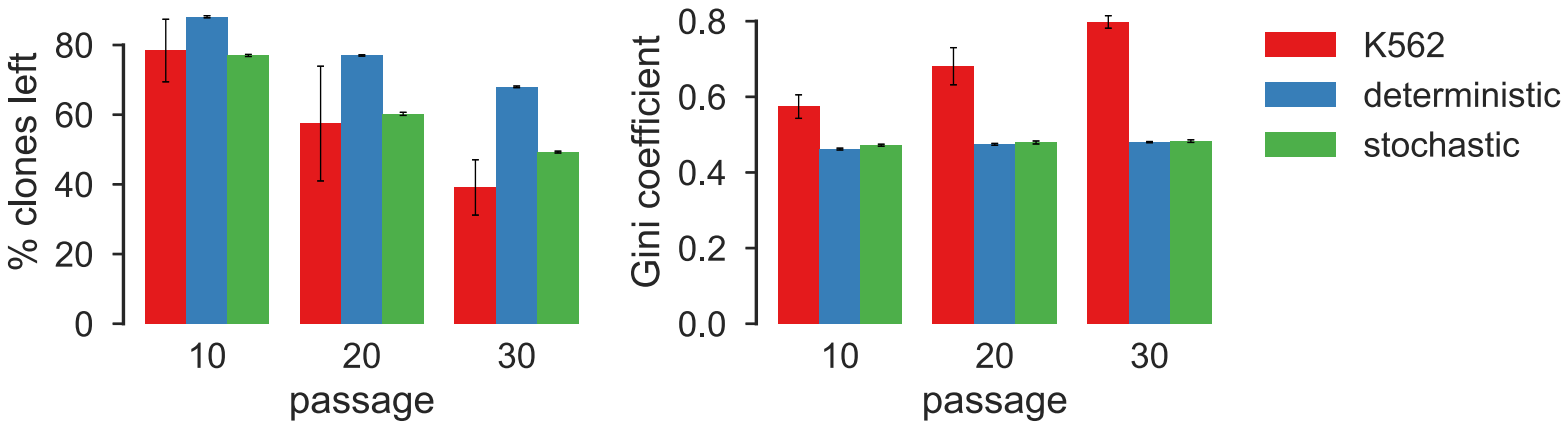
COMPUTATIONAL MODEL OF SIMPLE GROWTH & PASSAGE



- Initialization
  - ~12.000 clones with size c_i and $\sum_i c_i = 3 \cdot 10^5$.
  - Clone sizes assigned too fit the experimental data.
- Growth
  - Each cell grows with a given rate r_i
  - Growth continues until $\sum_i c_i = 4 \cdot 10^6$
- Passage
  - $3 \cdot 10^5$ cells are taken randomly and passed to the next generation

SIMULATIONS WITH STOCHASTIC GROWTH & PASSAGE

- All cells growth with rate r
  - deterministic: $c_i(t + \Delta t) = c_i(t)e^{r\Delta t}$
  - stochastic: c_i evolves with τ-leaping Gillespie algorithm



- Clones disappear at a rate similar to that *in vitro*
- Clonal dominance does not develop

TEST HYPOTHESES FOR CLONAL DOMINANCE

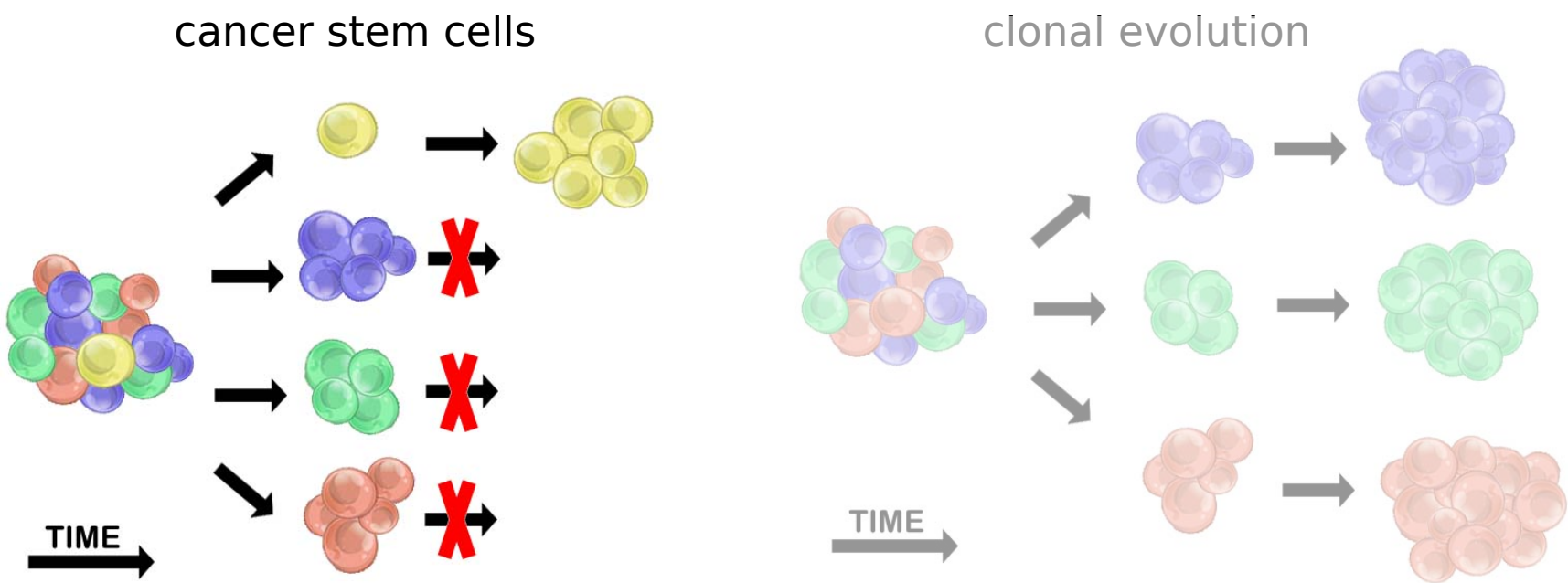
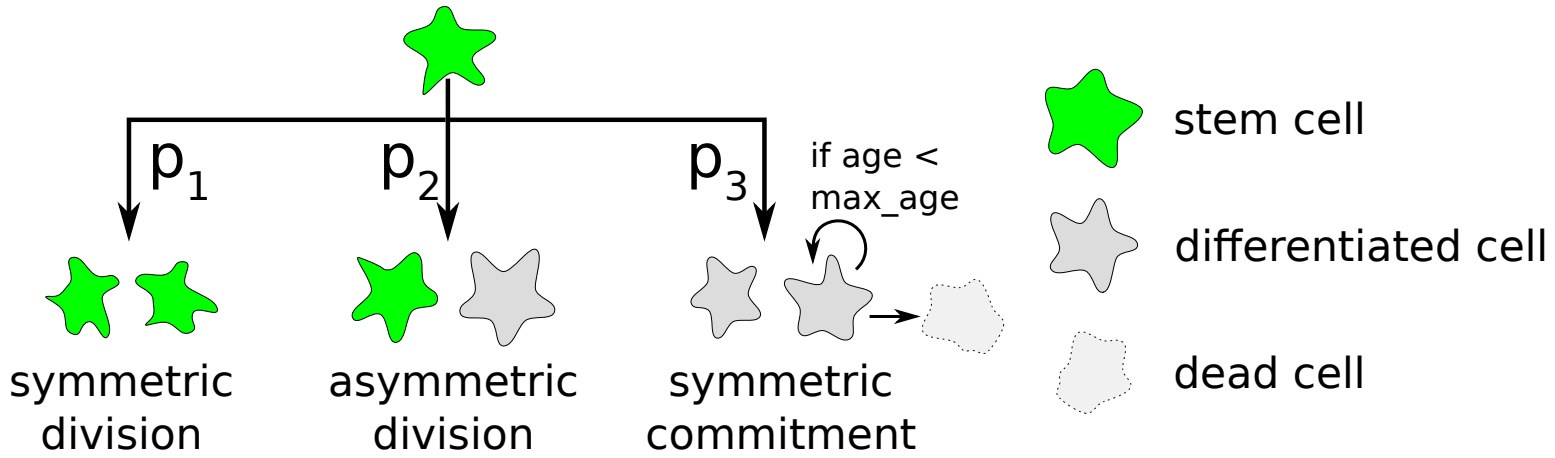


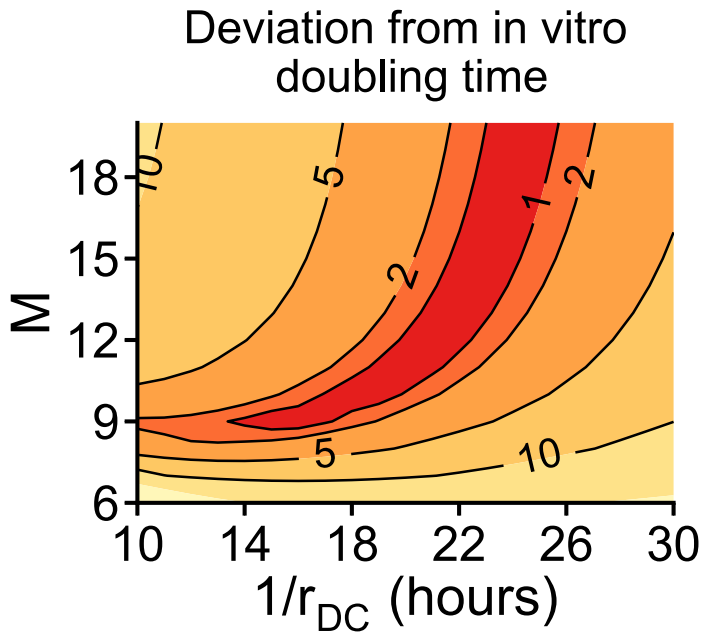
Image adapted from https://en.wikipedia.org/wiki/Tumour_heterogeneity

MODEL WITH CANCER STEM CELLS

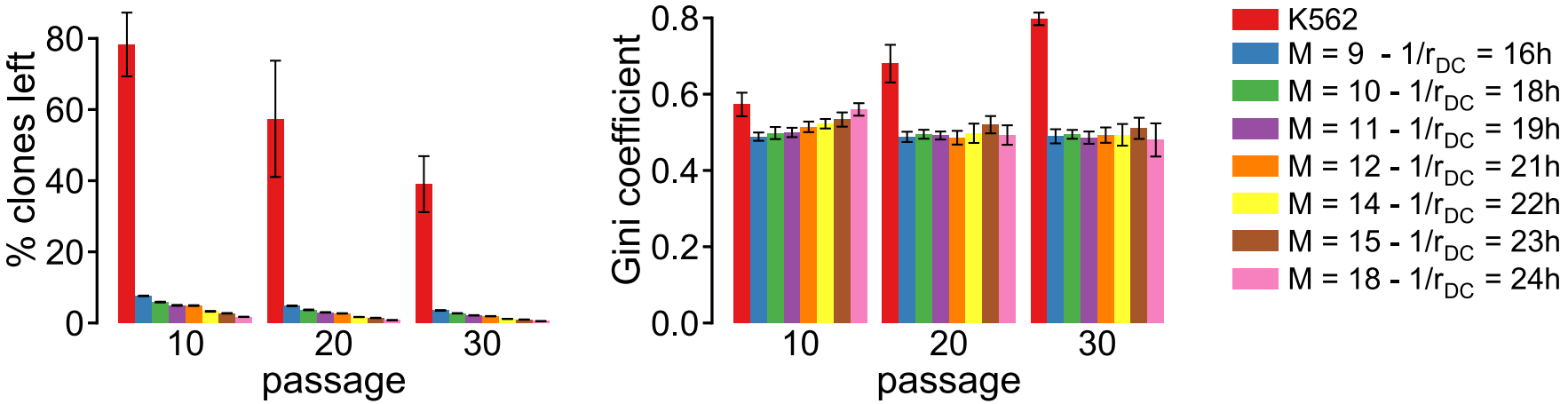


model based on Weekes *et al.*, Bull. Math. Biol., 2014

- Parameterization based on analytical solution
- Monotonic growth for $p_1 > p_3$
- Population growth rate depends on r_{DC} and maximum DC age (M)

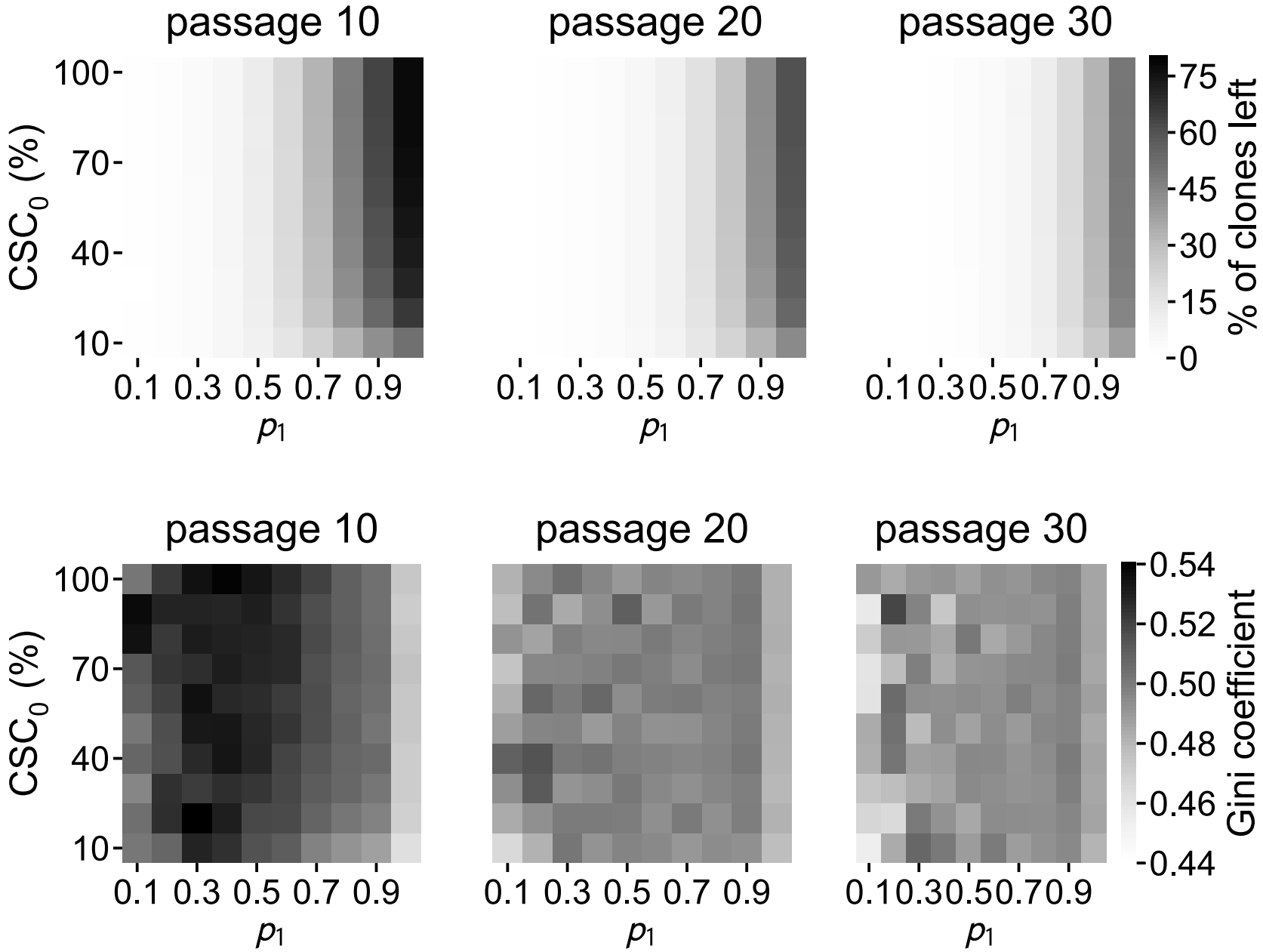


MODEL WITH CANCER STEM CELLS



- Cancer stem cells do not induce clonal dominance
- Almost all clones disappear

MODEL WITH CANCER STEM CELLS



TEST HYPOTHESES FOR CLONAL DOMINANCE

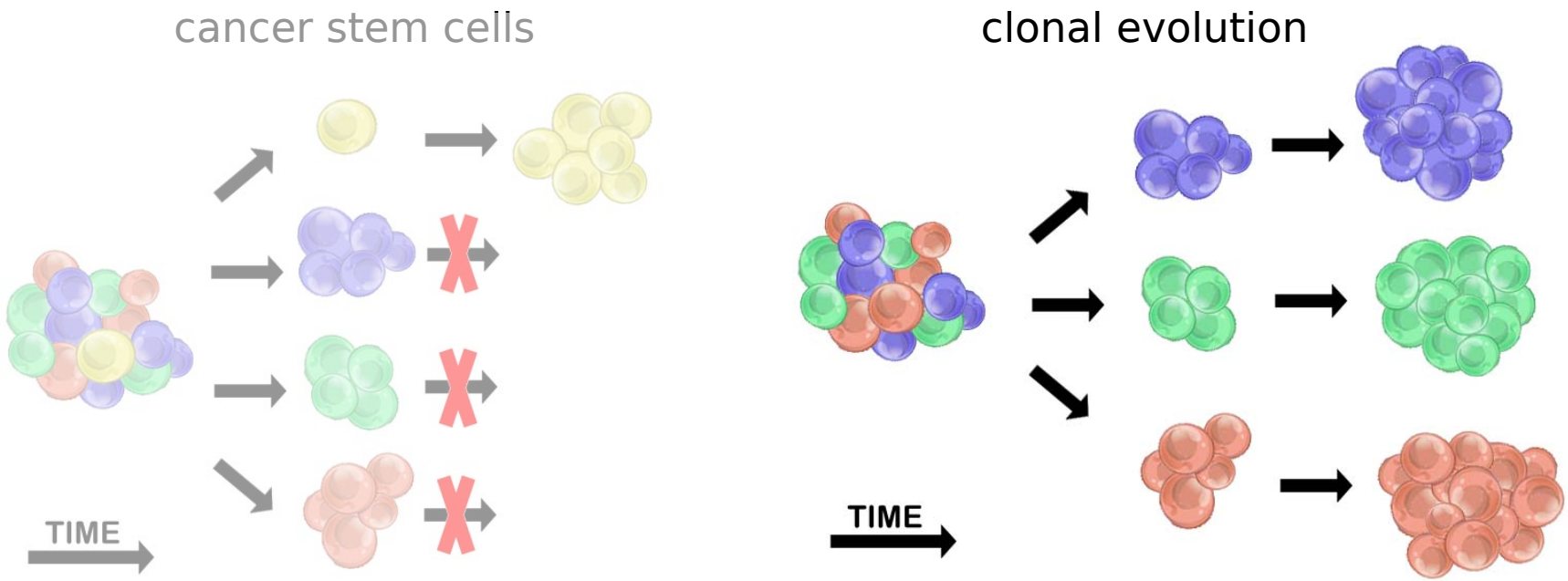
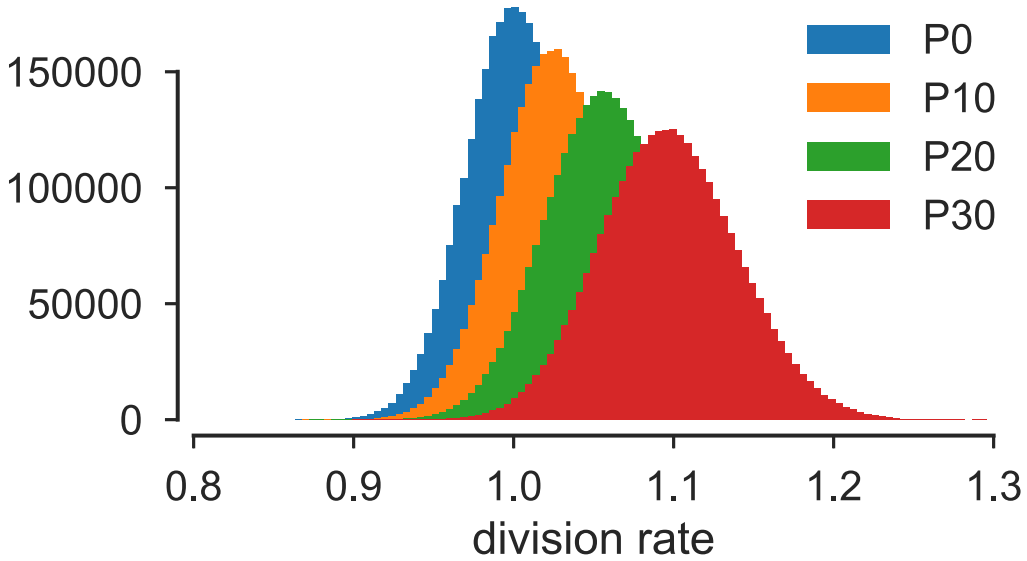


Image adapted from https://en.wikipedia.org/wiki/Tumour_heterogeneity

- Clonal evolution: division rate mutation
- Mutation requires tracking of individual cells

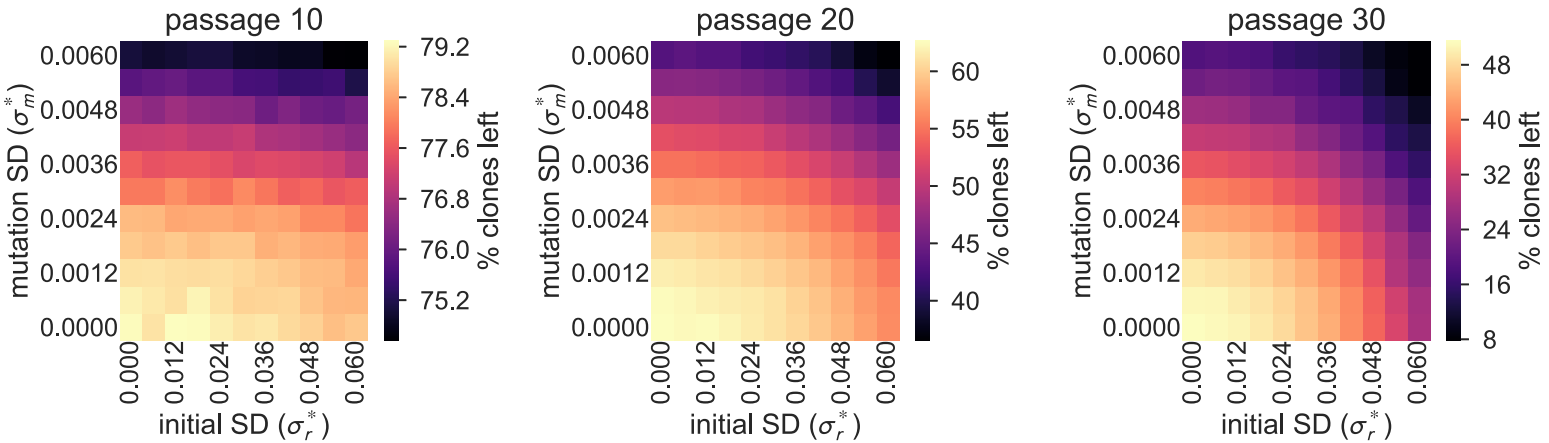
AGENT BASED MODEL (ABM) WITH WITH CLONAL EVOLUTION

- Cell i has a division rate r_i and a barcode.
- Initial variation to mimic evolution before experiment:
  - $r_i = rY$ with Y taken from $\mathcal{N}(1,\sigma_r^2)$
- Division
  - division rate mutates: $r_{\text{child}} = r_{\text{parent}} X$ with X taken from $\mathcal{N}(1,\sigma_m^2)$
- Division rate increases during the experiment

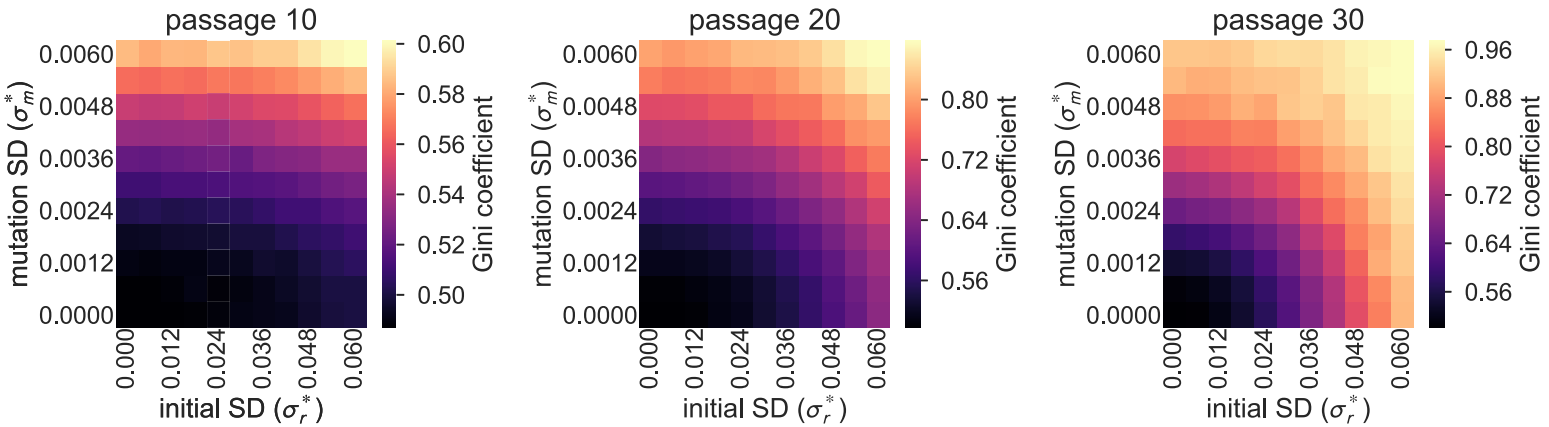


ITERATED GROWTH & PASSAGE WITH CLONAL EVOLUTION

- Clone loss increases with division rate variation

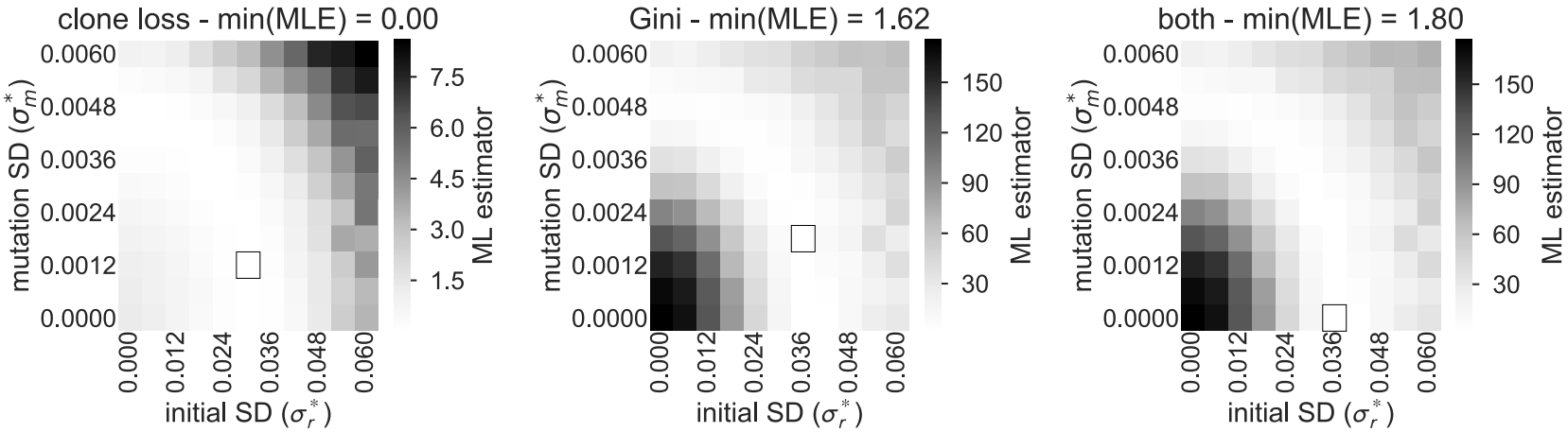


- Gini coefficient increases with division rate variation

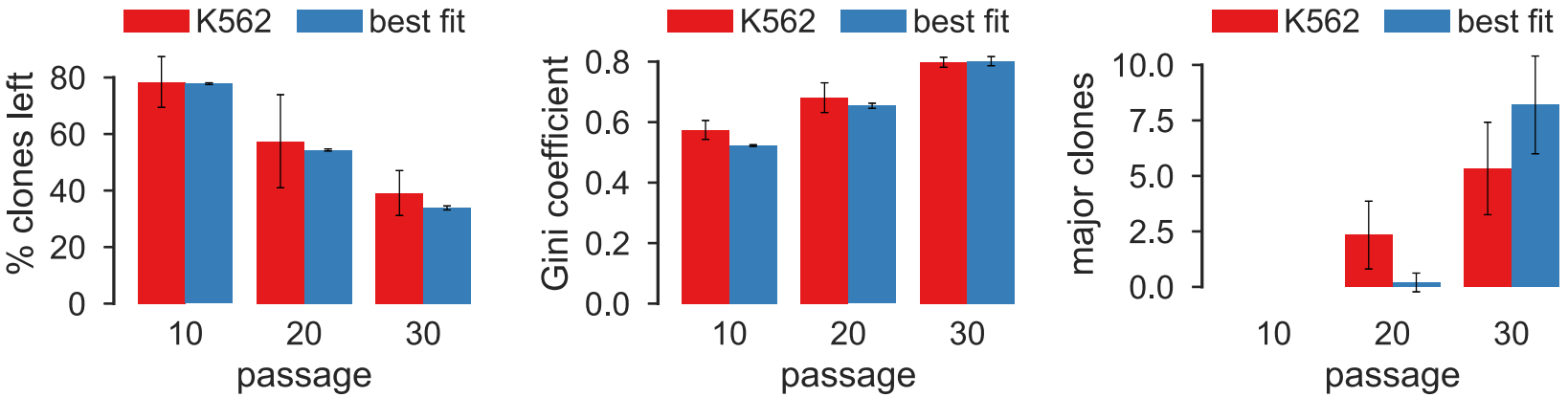


MATCHING ABM TO *IN VITRO* RESULTS FOR K562

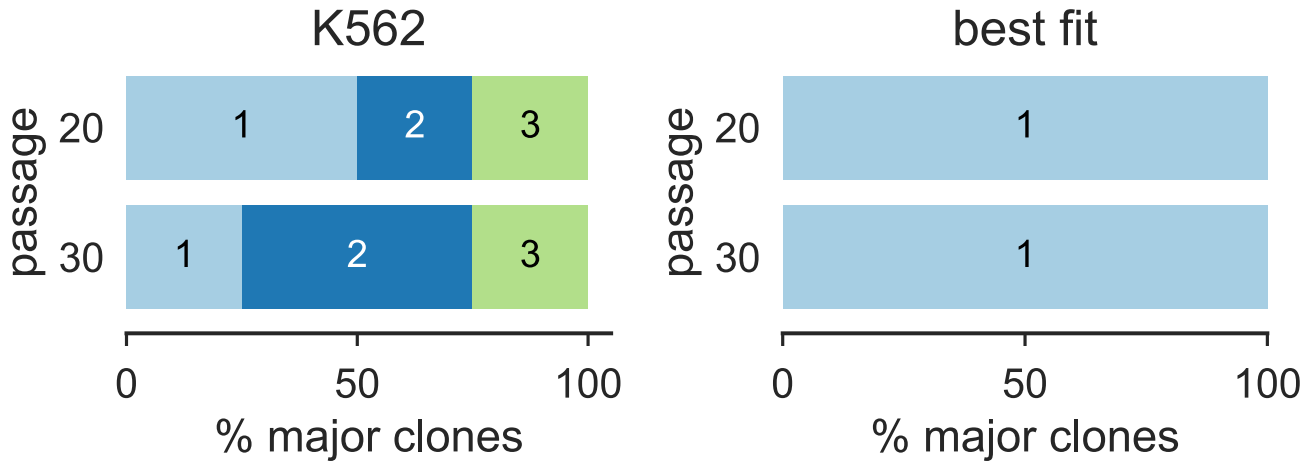
- Maximum likelyhood estimation that includes the 3 time points



- Results for best fit for Gini coefficient



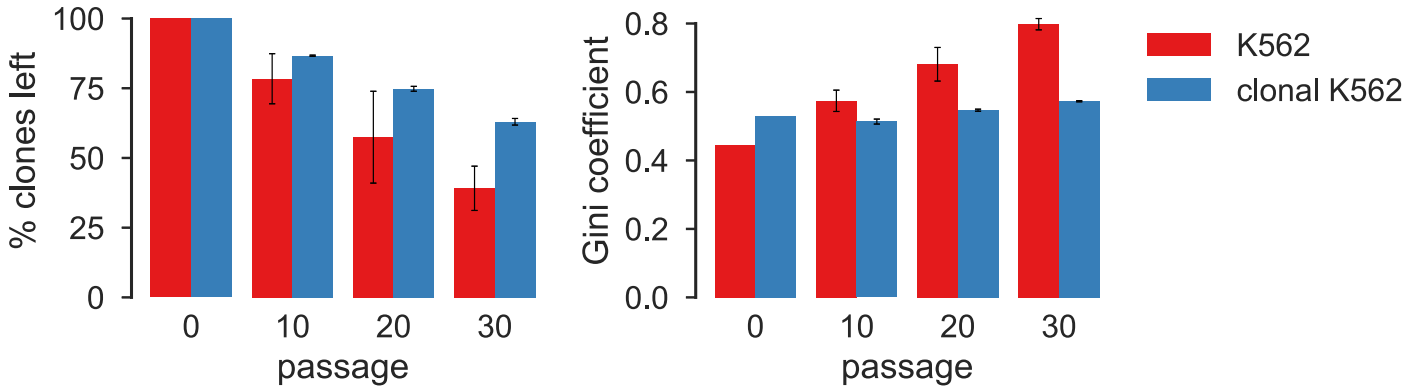
MODEL DOES NOT REPRODUCE CLONAL OVERLAP



- No clonal overlap in the simulation because of initialization:
  - Simulation initialization: large population with clones distributed based on data at P0
  - Actual initialization: cells growth for 7-8 days after barcodes are inserted
- **There should be correlation between division rates for cells of the same clone**

IN VITRO RESULTS FOR CLONAL K562

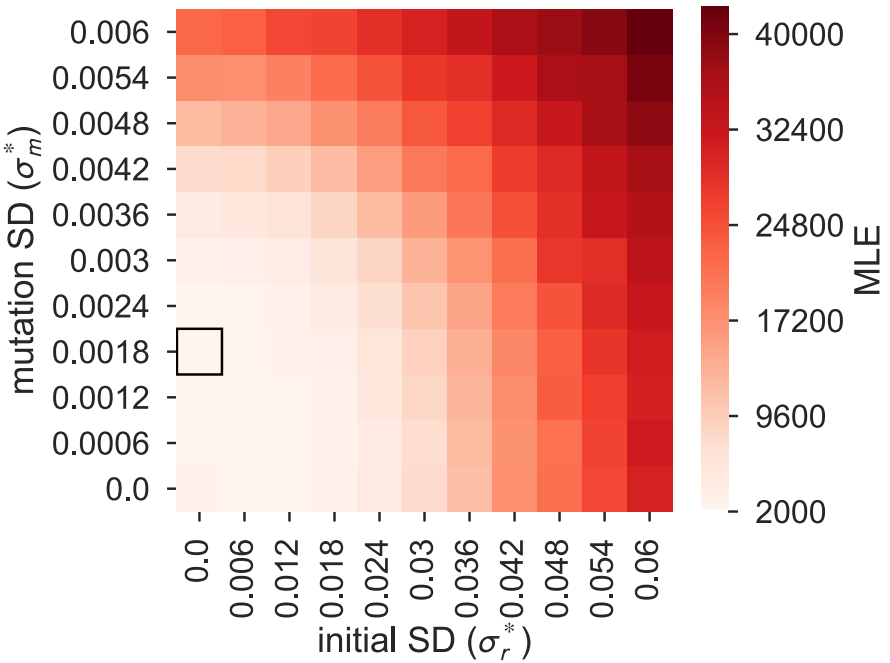
- clonal K562 cell line is derived from a single cell



- Less clone loss compared to K562 cell line
- No development of clonal dominance

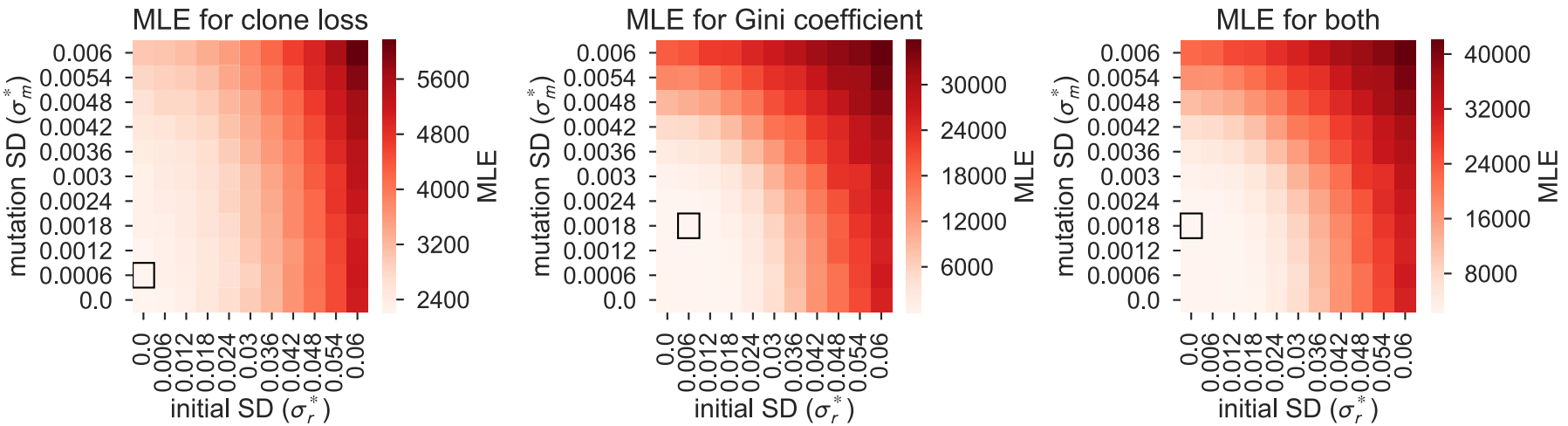
MATCHING ABM TO CLONAL K562 RESULTS

- Expectations:
  - Less initial variation ⇒ lower σ_r than for K562
  - Same cell type ⇒ similar σ_m as with K562
- Changes as expected:

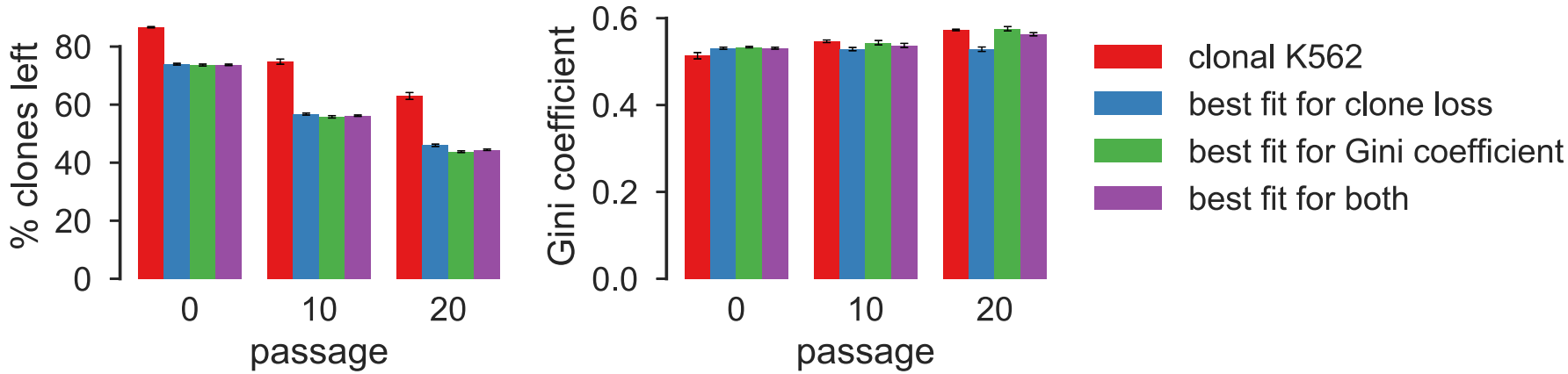


	clonal K562	K562
σ_r	0	0.036
σ_m	0.002	0.0018
min(MLE)	~2000	~5

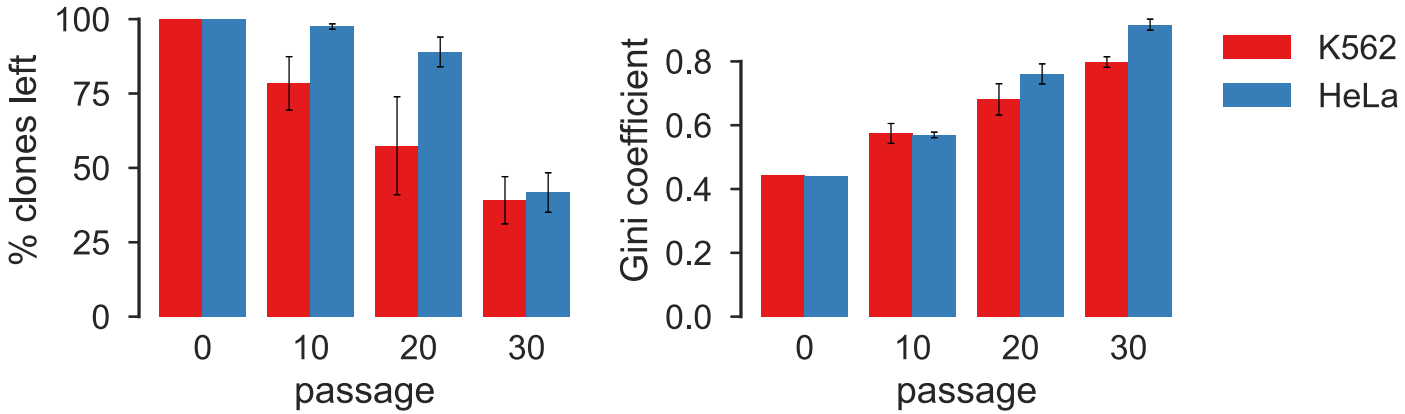
MATCHING ABM TO CLONAL K562 RESULTS



- Both clone loss is hard to match (min(MLE) = ~2000)
- Gini coefficient matches better (min(MLE) = ~5)

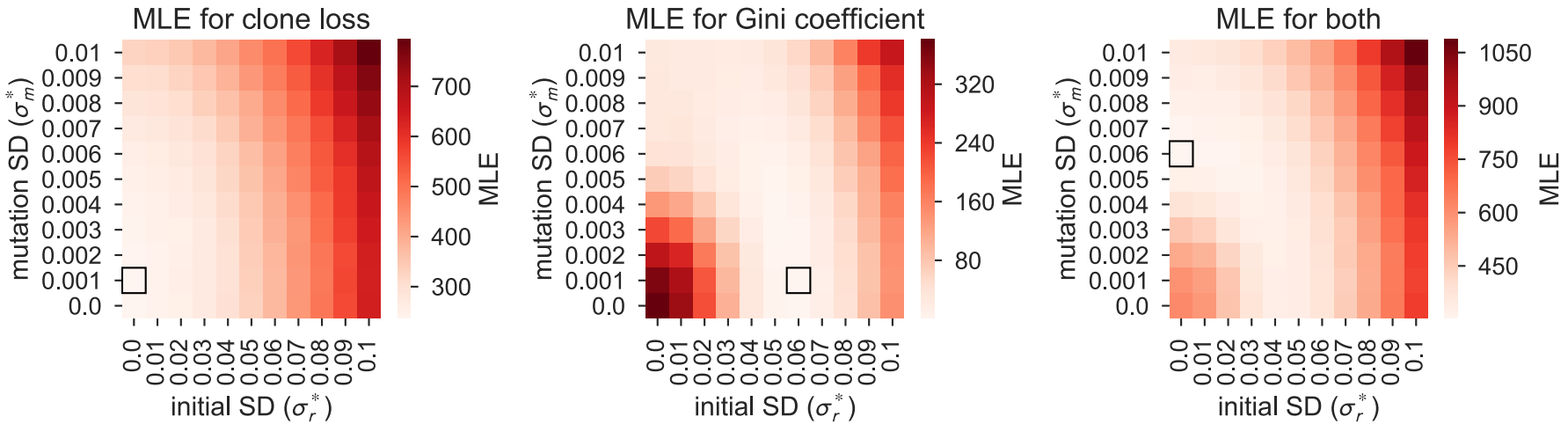


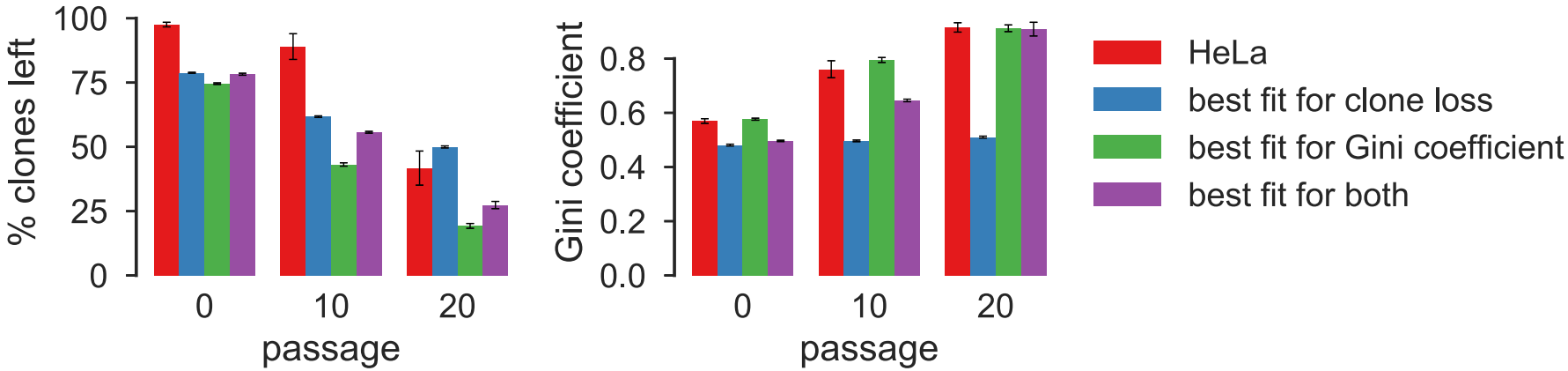
IN VITRO RESULTS FOR HELA CELLS



- Clone loss starts late
- Clonal dominance develops similar to K562 cell line

MATCHING ABM TO HELA RESULTS





Better fit for Gini coefficient

CONCLUSION

- Model based on cancer stem cells does not match *in vitro* iterated growth and passage
- Model based on clonal evolution can match *in vitro* iterated growth and passage
  - Model fits well to changes in clone size distribution observed *in vitro*
  - Model predicts same mutation dynamics for polyclonal and monoclonal K562 cells
  - Model fails to predict correct clone loss for monoclonal K562 and HeLa cells
  - Model fails to reproduce major clone overlap

NEXT STEP - SPACE

- *In vivo* lineage tracing shows various clone size dynamics

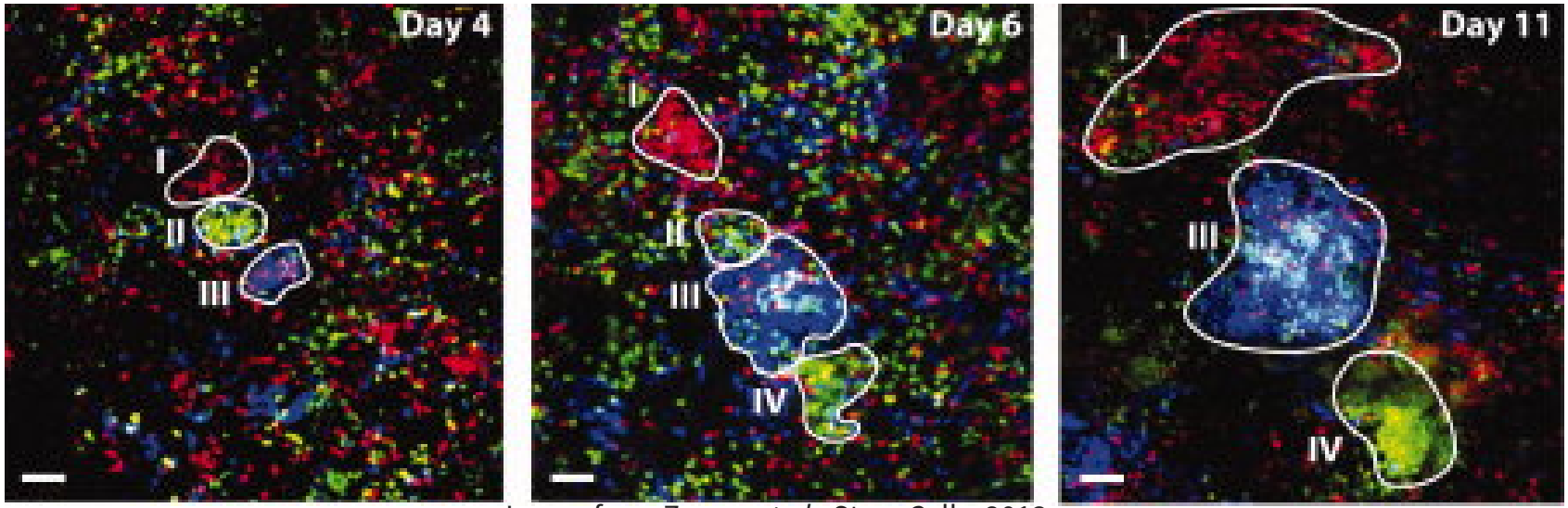


Image from Zomer *et al.*, Stem Cells, 2013

- Current model cannot simulate *in vivo* tumor development
- Matthijs should be able to tell us more in ~6 months....

FINALLY

- Accepted for PLoS Computational Biology
- It took
  - 6 submissions
  - to 5 journals
  - over ~14 months

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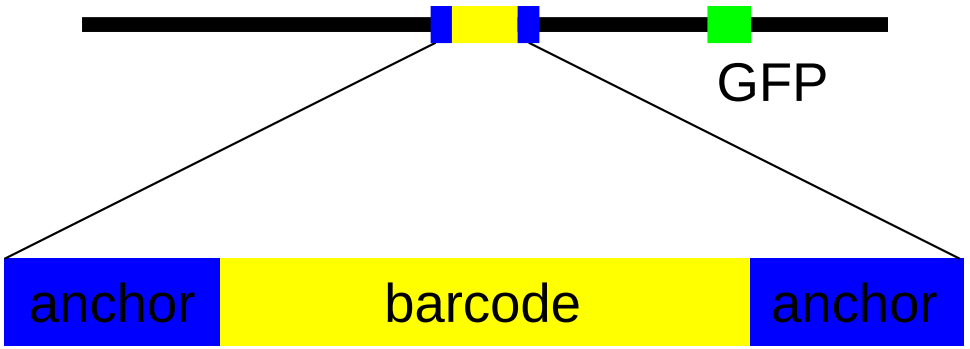
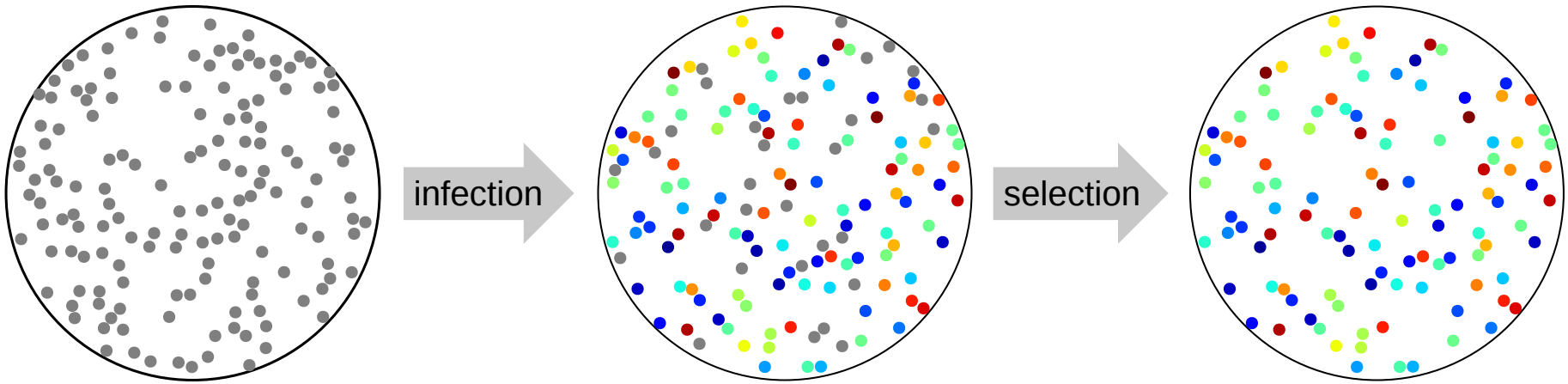
LINEAGE TRACING WITH GENETIC BARCODES

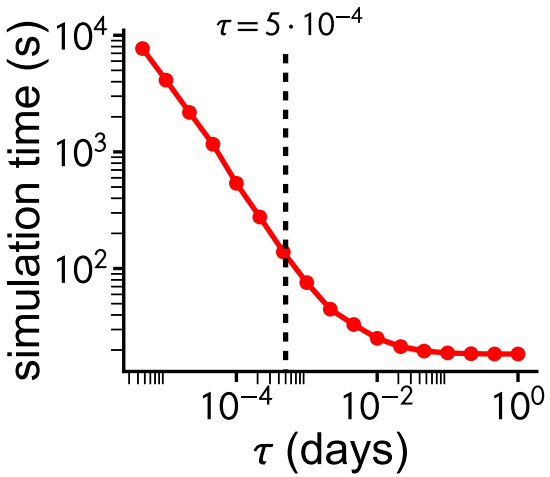
- construct with random base pair sequence

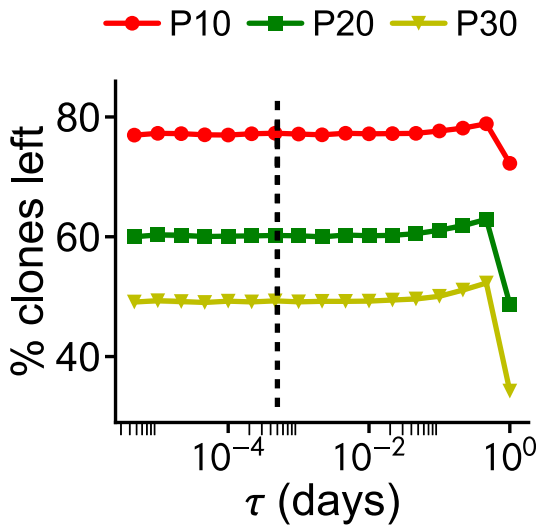


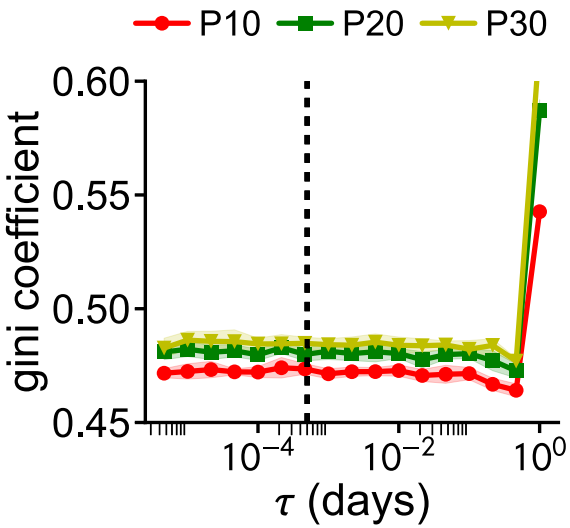
- construct is inserted, e.g. with a virus vector



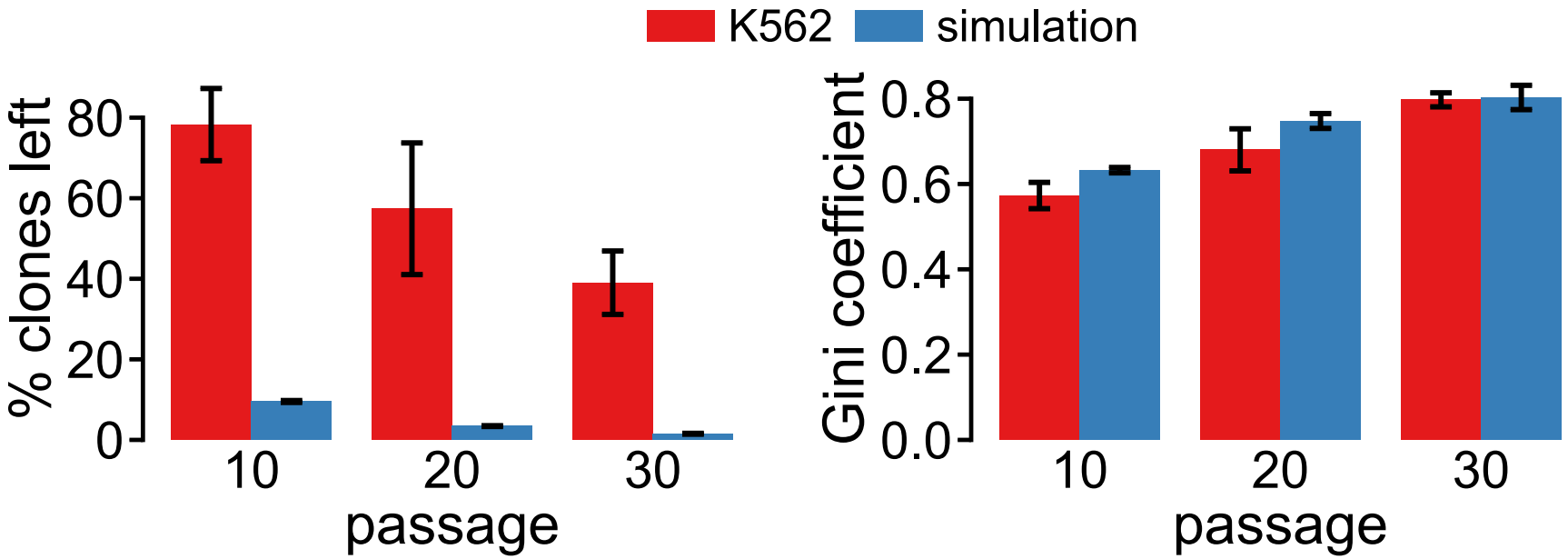
TAU-LEAPING INTERVAL







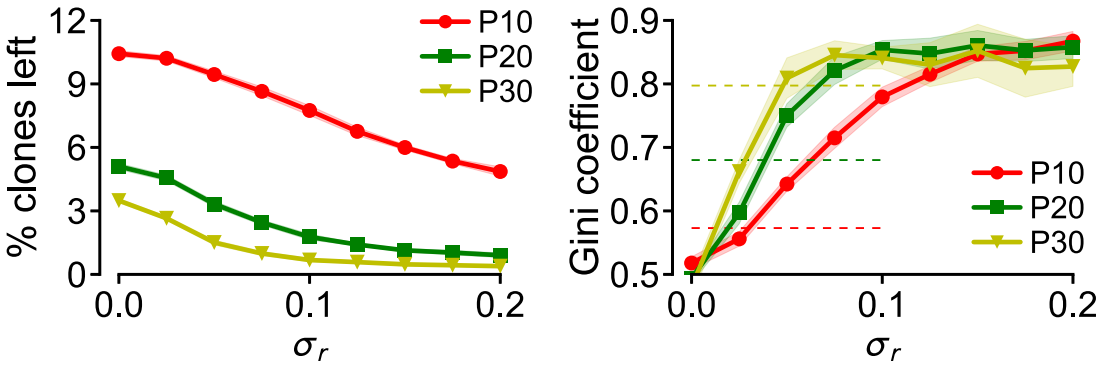
MODEL WITH CSC AND DIVISION RATE HETEROGENEITY



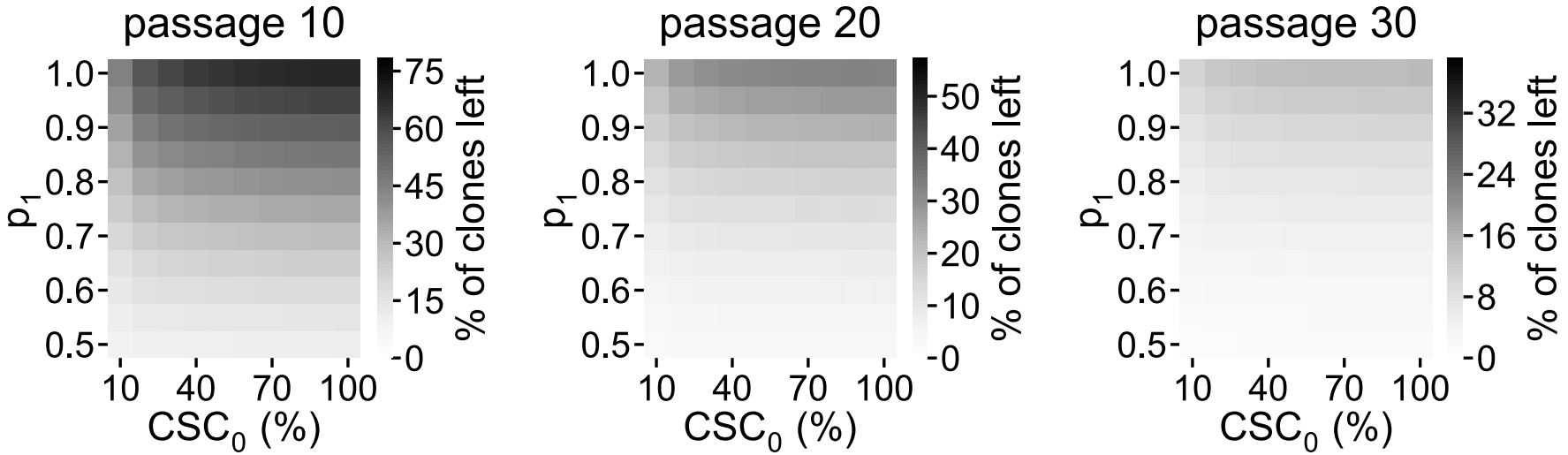
Division rate heterogeneity induces clonal dominance, but does not reduce excessive clone loss

MODEL WITH CSC AND DIVISION RATE HETEROGENEITY

- Division rate standard deviation determines clonal dominance

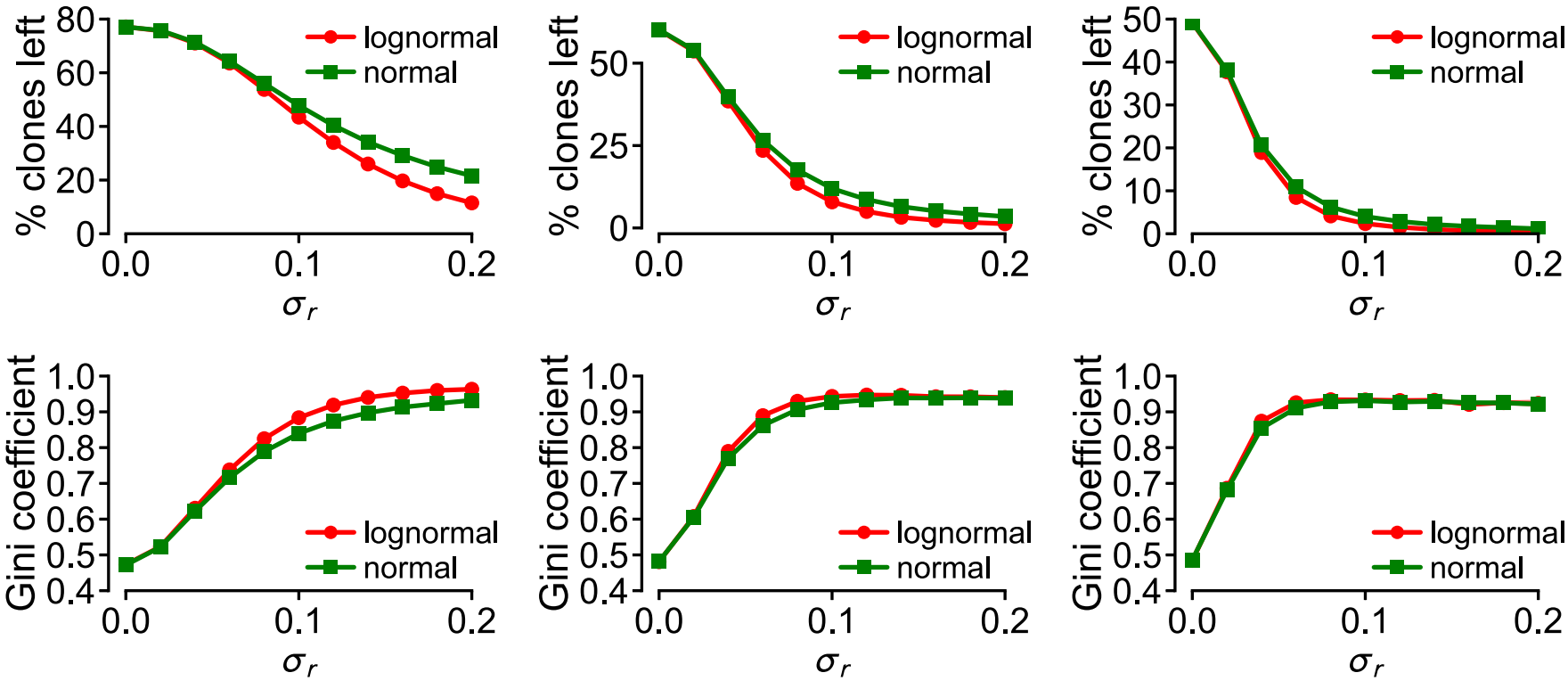


- CSC_0 and p_1 determine clone loss



Best fit with 100% CSCs that only divide into CSCs.

ALTERNATIVE DIVISION RATE DISTRIBUTION



CLONAL K562 CLONE LOSS CANNOT BE MATCHED

- Minimal clone loss for a model without any division rate variation
- Clone loss observed with clonak K562 is larger than that in a simulation without division rate variation