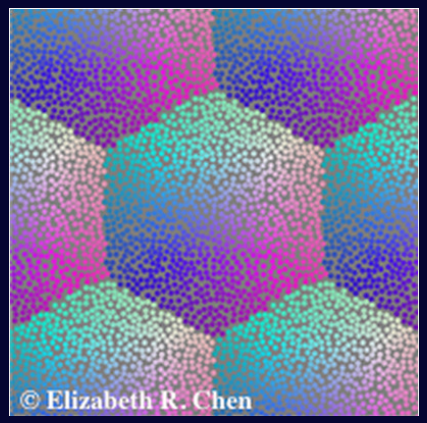
stochastic effects in hematopoiesis

Jorge M. Pacheco
Math – U. Minho

atpgroup
http://www.ciul.ul.pt/~ATP/



21-FEB-2013

If cells play dice, can we gamble our way out of cancer ?

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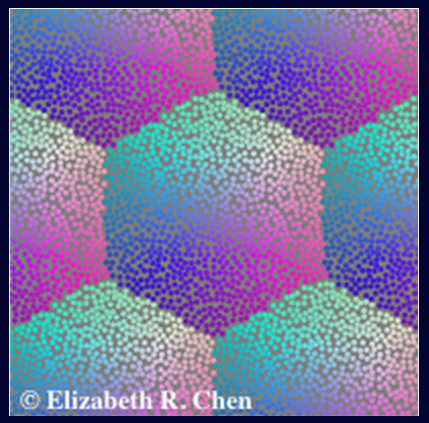


21-FEB-2013

Cancer

(a poor summary of some of David Dingli's slides)

- cancer is a consequence of multicellularity
- cellular genome is under permanent attack (environmental or metabolic genotoxic agents)
- DNA replication machinery is not perfect

} mutations

- many mutations are neutral
- others → malignant tranformation → clonal development
- impact of mutations: μ rate, # cells@risk, cell-lifetime

tissue architecture

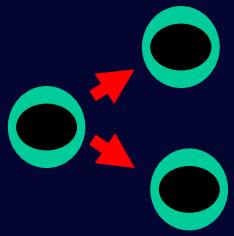
- tissue architecture has evolved
- most tissue cells have a ↘ lifetime & a ↗ turnover

  → minimize impact of mutations
- many tissues evolved a hierarchical structure

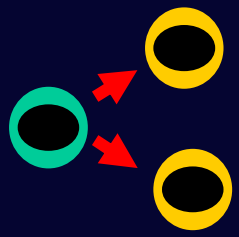
  → tree-like structure
- at the root of the tree are the tissue-specific stem-cells

- example: hematopoiesis
- stem-cell concept was developed in hematopoiesis and has been extended to many other tissues
- *HSC resilience* relies on ↘ # & ↘ turnover of stem cells

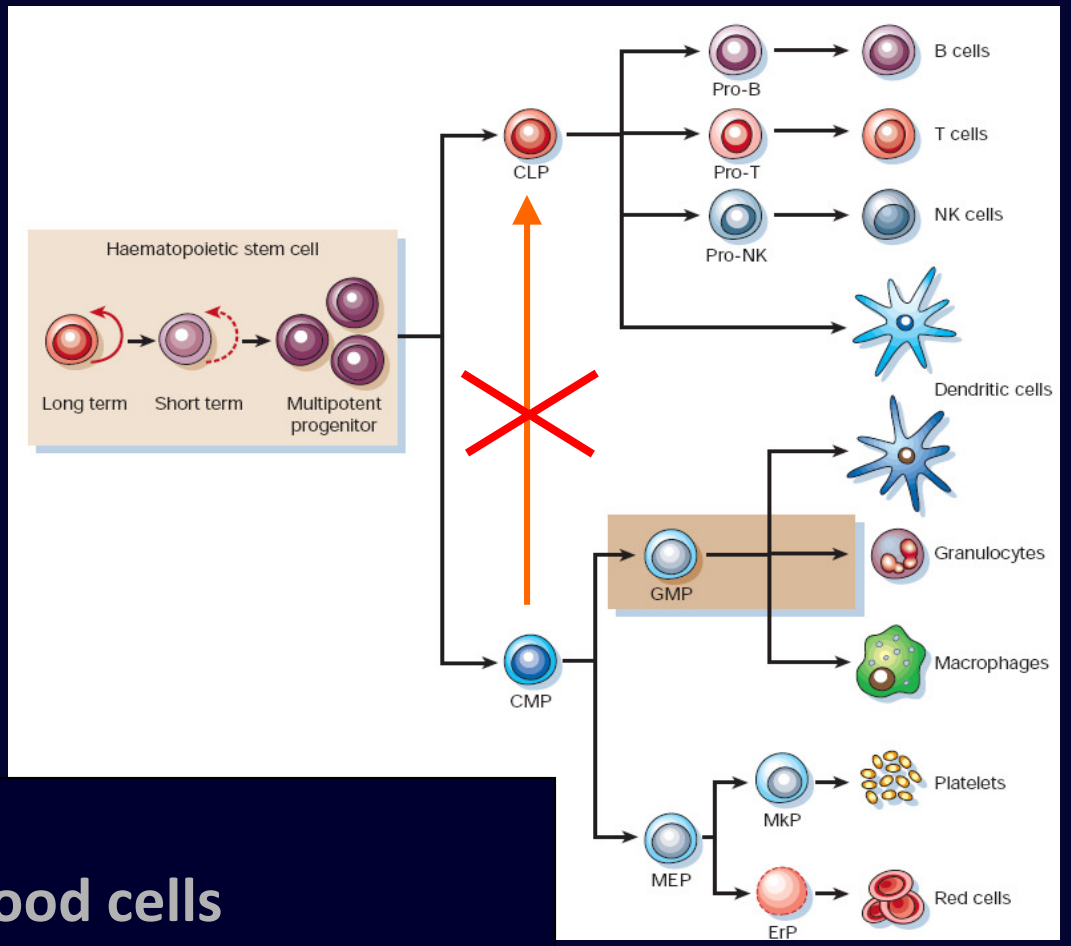
hematopoietic stem cells (HSC)



self-renewal :
capacity to clone
themselves

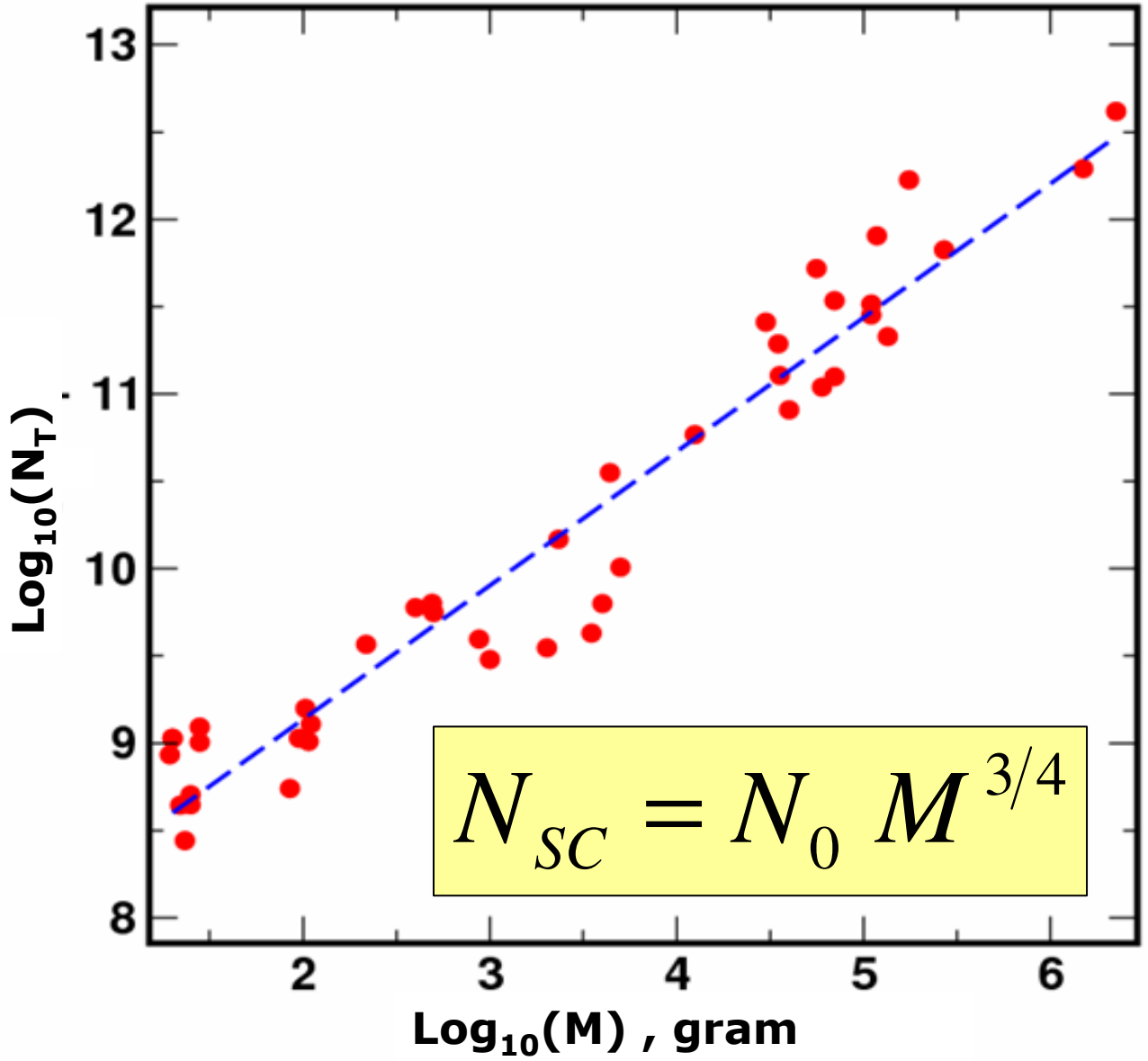


differentiation :
capacity to differentiate
into all other types of blood cells



stemness** is a matter of degree – you have to stand at the **root** of the **hematopoietic tree

allometric scaling of hematopoiesis in land mammals



Dingli & Pacheco, *PLoS ONE*, 2006

allometric scaling of hematopoiesis in land mammals

use experimental estimates for cats for calibration (fix N_0):
under normal conditions, ≥ 40 ! (*Abkowitz et al, Blood, 2002*)

what	model predictions	×	experimental data
HSC in humans *cat = 40*	**385**		**~400** *(Buescher et al, J Clin Invest, 1985)*
rate HSC division *cat post-TRX = 8 week^{-1}*	**60 week^{-1}**		**~ 52-104 week^{-1}** *(Rufer, et al, J Exp Med, 1999)*
human post-transplant *cat = 13*	**111**		**~ 116** *(Nash et al, Blood, 1988)*
mouse	**1**		**1** *(Abkowitz et al, PNAS , 1995)*
rate macaques	**23 week^{-1}**		**23 week^{-1}** *(Shepherd et al, Blood , 2007)*
rate baboons	**36 week^{-1}**		**36 week^{-1}** *(Shepherd et al, Blood , 2007)*

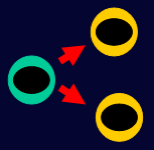
Dingli & Pacheco, *PLoS ONE*, 2006

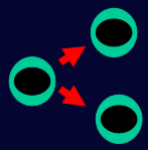
the hematopoietic *tree*

- *in humans ~ 400 HSC divide each once per year*
- *but : daily output of bone marrow ~ 3.5 x 10^{11} cells !!!*

> ***how to explain this enormous amplification given the slow replication rate of HSC ?***

- *one must consider :*

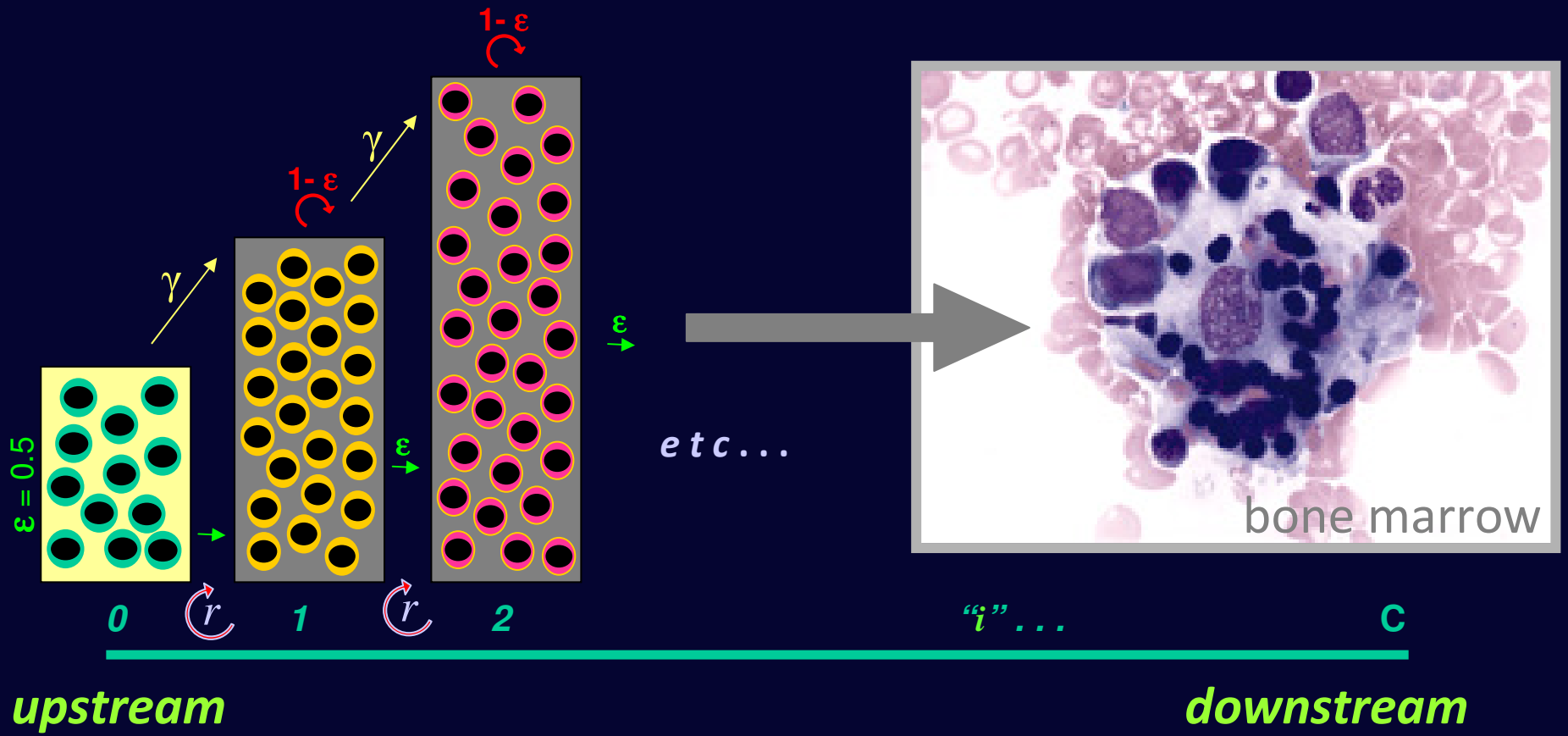
differentiation  ε

amplification  1- ε

asymmetric division : more parameters, see Dingli et al. *PLoS-CB*, 2007

the hematopoietic *tree*

- *we consider a compartmentalized structure in which cells from upstream compartments flow into downstream compartments , under stationary flux conditions;*



Dingli, Traulsen & Pacheco, *PLoS-ONE*, 2007

deterministic dynamics of the hematopoietic *tree*

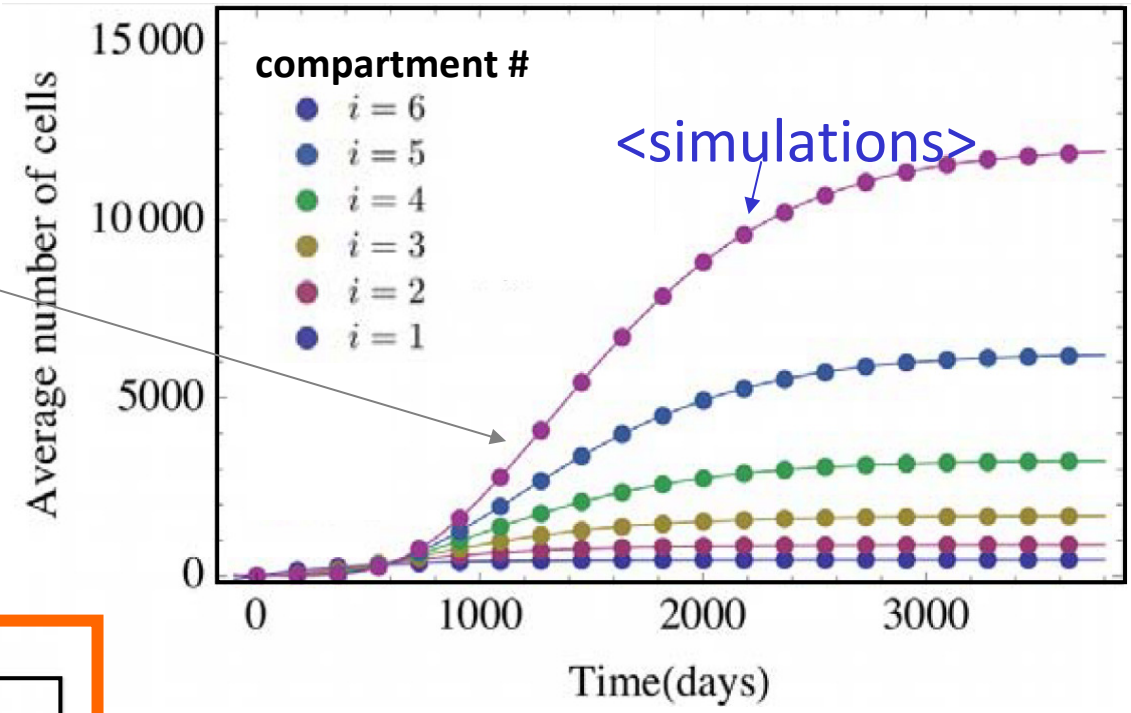
$$\dot{N}_i(t) = -d_i N_i(t) + b_{i-1} N_{i-1}(t)$$

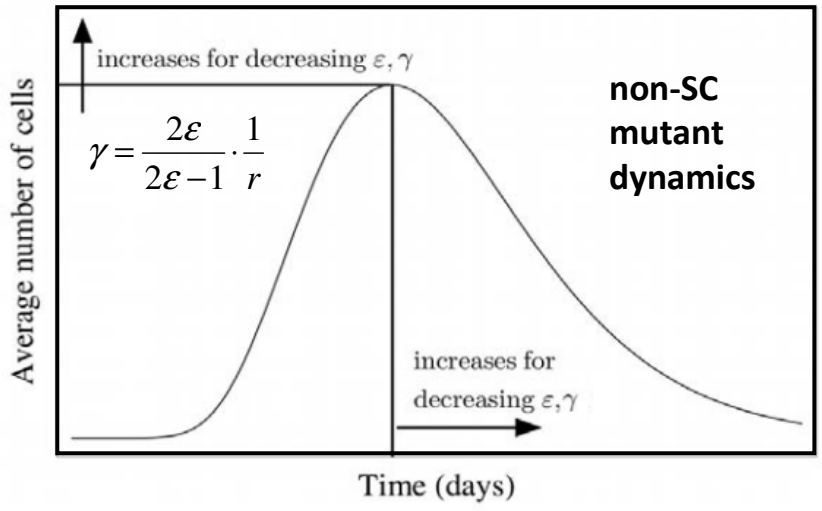
$$d_i = (2\varepsilon - 1) r_i \qquad r_i = r^i r_0$$

$$b_i = 2\varepsilon r_i$$

analytic solution

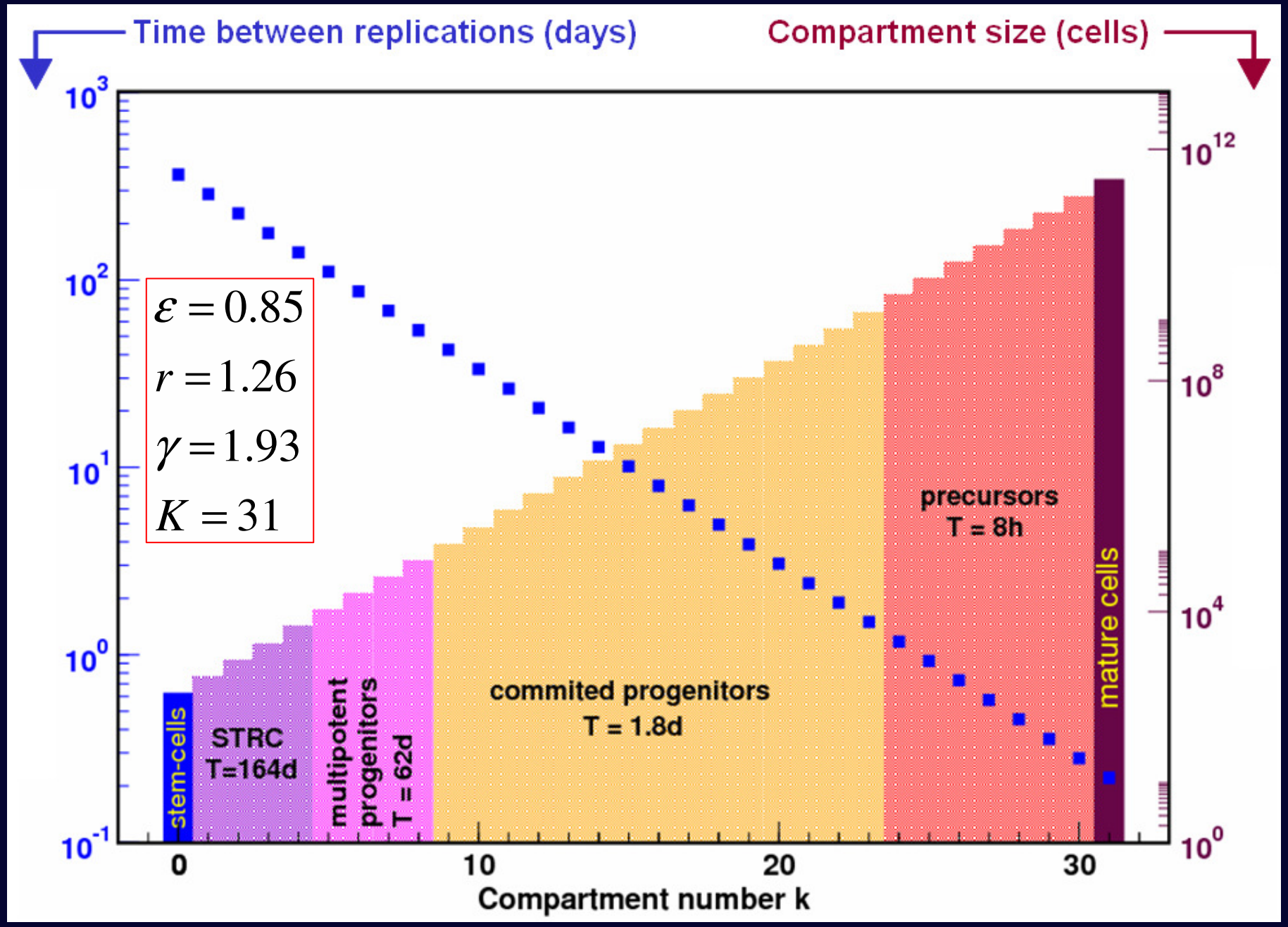
Werner *et al.*, PLoS-CB, 2011





the deterministic model provides numerous insights onto the average dynamics in the hematopoietic tree; for instance, that even if you kill the *LSC*, you may still die from its progeny (ex: CML)

the hematopoietic *tree*



Dingli, Traulsen & Pacheco, *PLoS-ONE*, 2007

DISEASE

The Economist, 13th September, 2008
(article on cancer stem cells)

scaling relations . . .

❖ *number of **HSC** in adult mammals :*

$$N_{SC} \approx 16.55\, M^{3/4}$$ [--]

❖ *number of **HSC** during human ontogeny :*

$$N_{SC} \approx 5.5\, m(t)$$ [--]

❖ ***HSC** replication rate :*

$$r_0 \approx 2.9\, M^{-1/4}$$ [*year*$^{-1}$]

❖ *average life-span of organism :*

$$L \approx 8.6\, M^{1/4}$$ [*year*]

([M] = kg)

simple implications . . .

***Hayflick hypothesis* (*1961*):**
cells undergo a limited number of divisions during their lifespan

from the scaling relations, each cell divides

$$N \sim rate \times lifespan \sim M^{-1/4} \times M^{1/4} \sim M^{0}$$

***that is, constant & independent of the mammalian species* :**

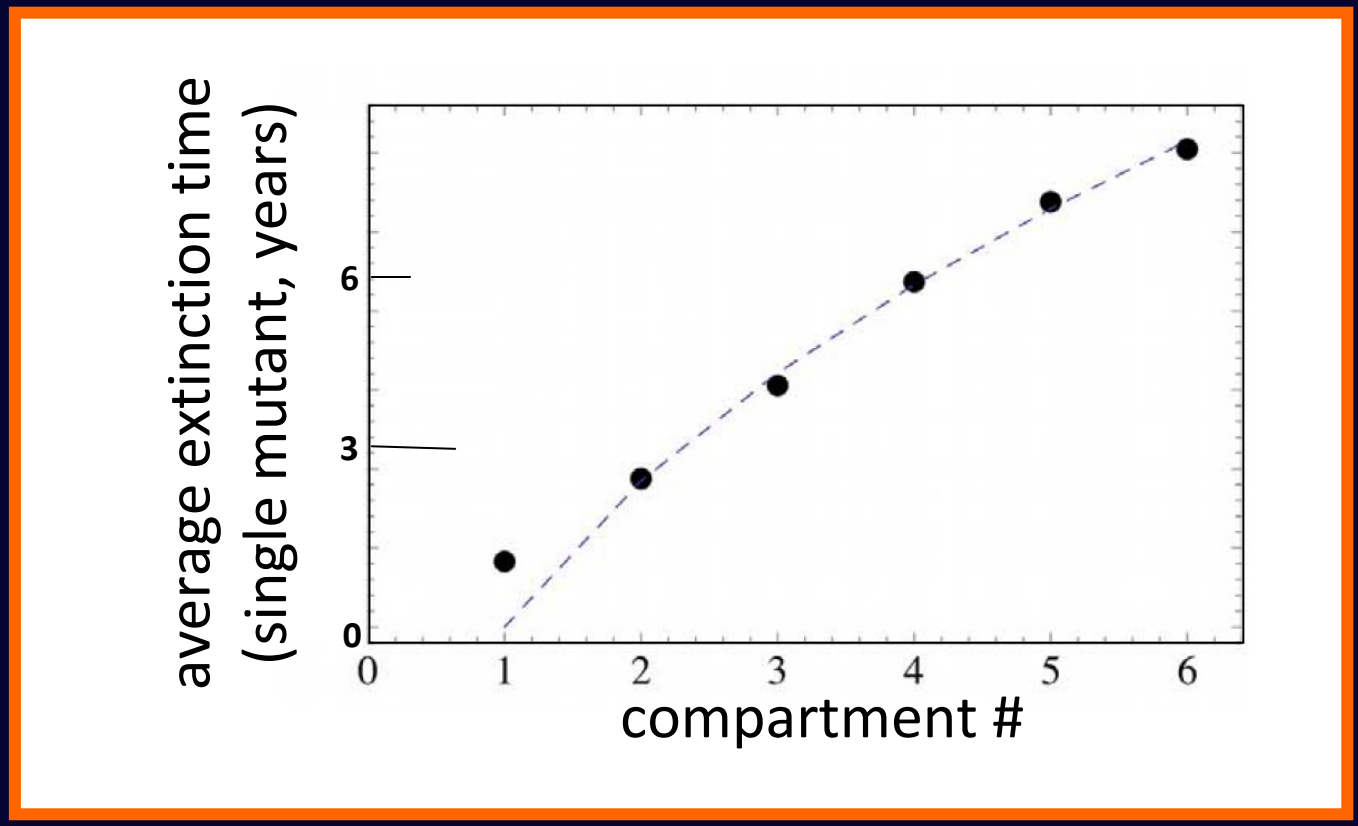
a mouse-HSC and an elephant-HSC replicate, on average, the same number of times during the ~2-year and the ~70-year lifespans of the mouse and elephant, respectively; humans are the exception, as we live much longer than lifespan estimate.

are stochastic effects important ?

- ***in vivo stochastic effects in hematopoiesis were found in 1996***

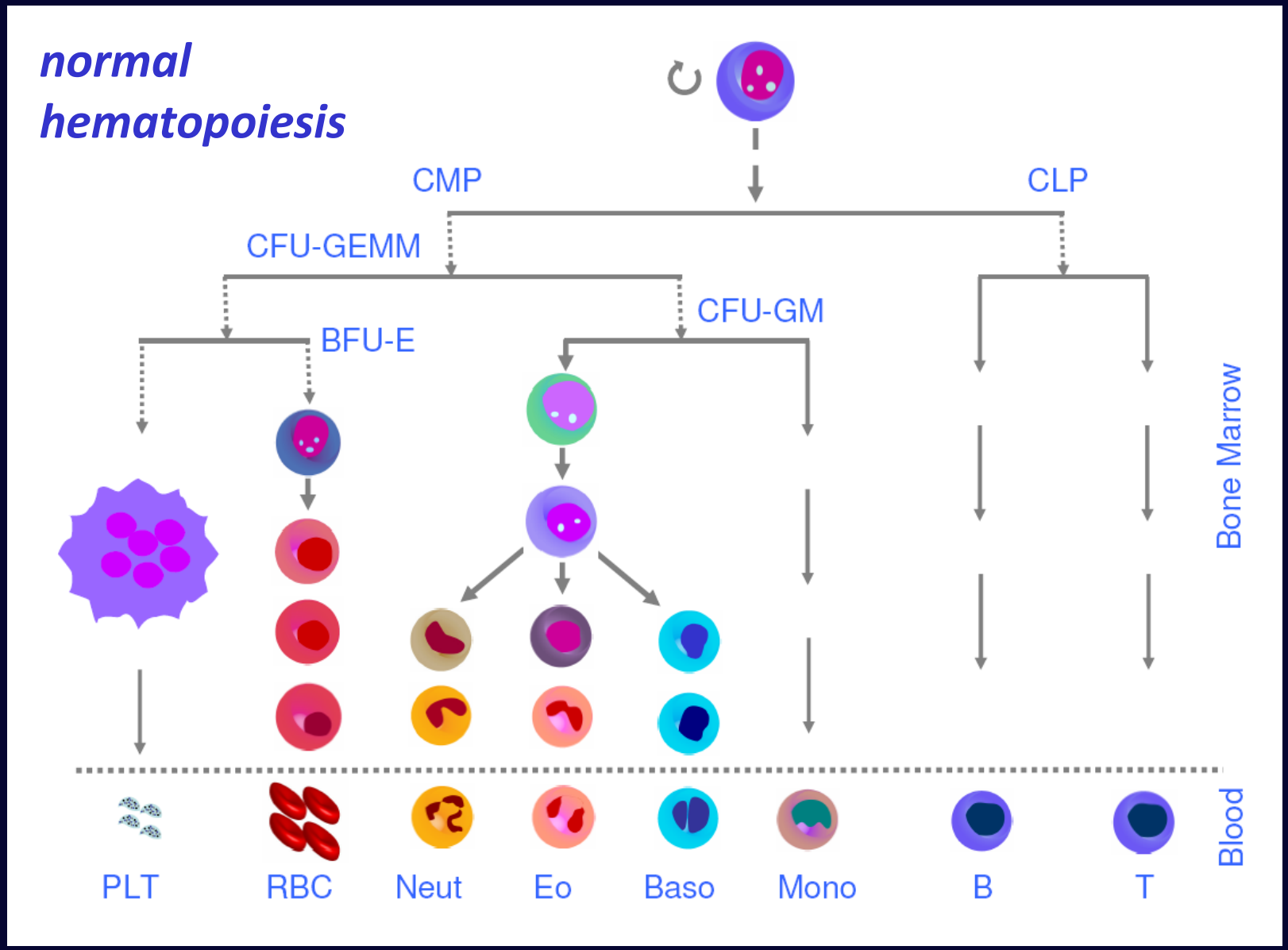
(*Abkowitz et al,* Nat. Med. , 1996)

- ***deterministic models (of hematopoiesis) at best describe average population dynamics behaviour, and may provide poor descriptions of small cell populations and neutral dynamics, in particular of HSCs ; this may have sizeable impact on disease dynamics***



trouble

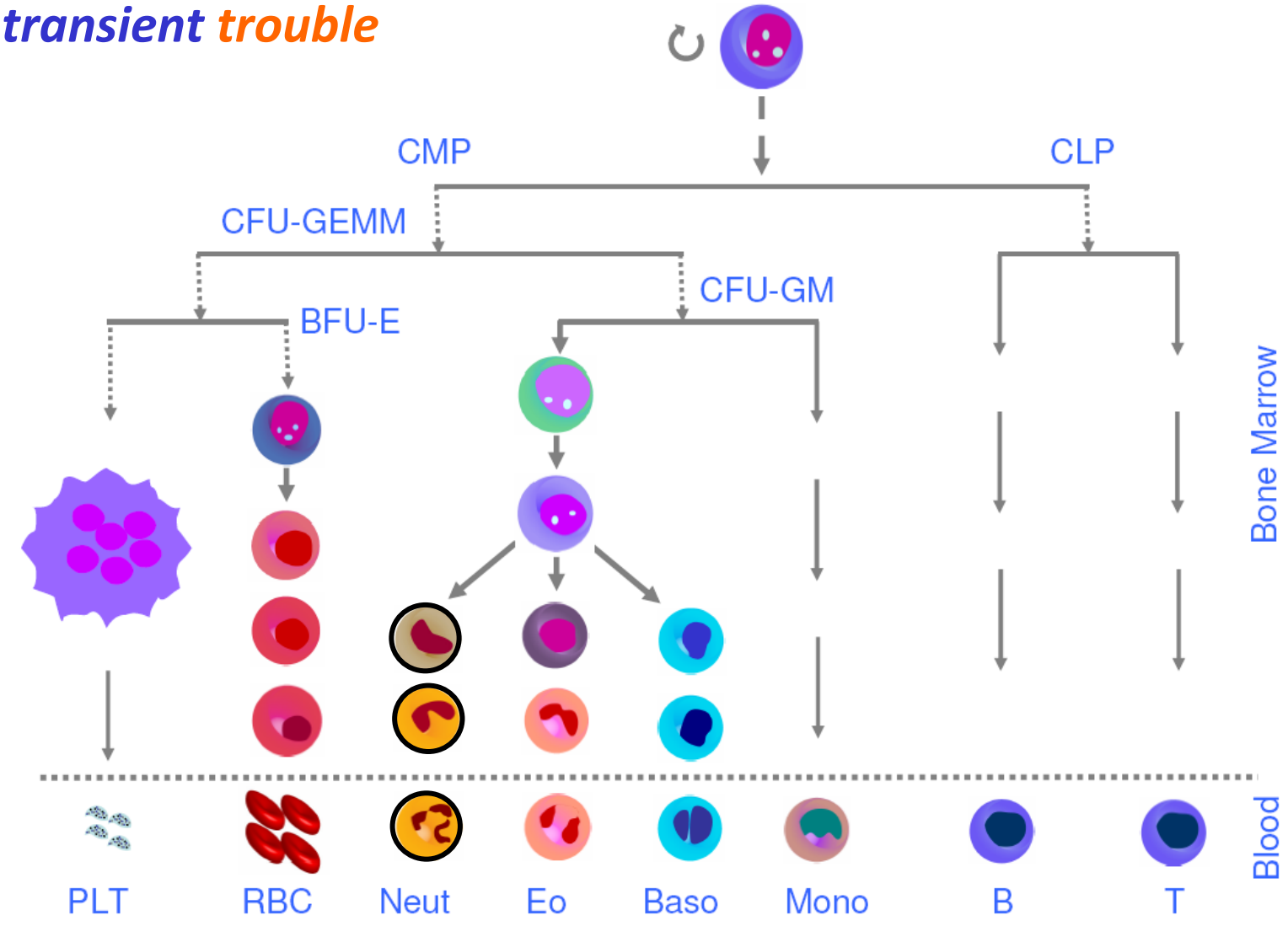
normal : $10^{-7} < \mu < 10^{-6}$ ***per cell per replication***



trouble

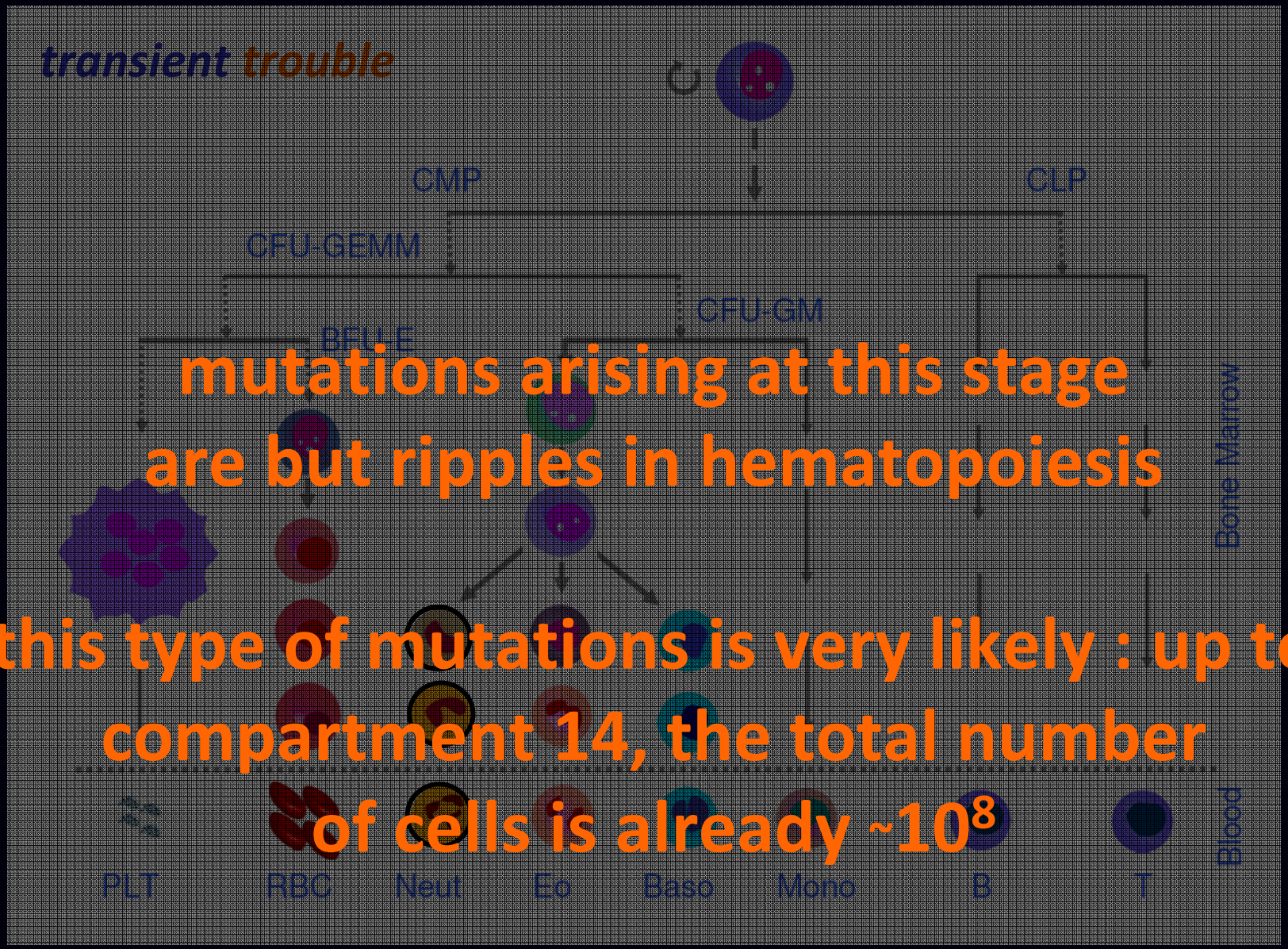
normal : $10^{-7} < \mu < 10^{-6}$ ***per cell per replication***

transient trouble



trouble

normal : $10^{-7} < \mu < 10^{-6}$ ***per cell per replication***

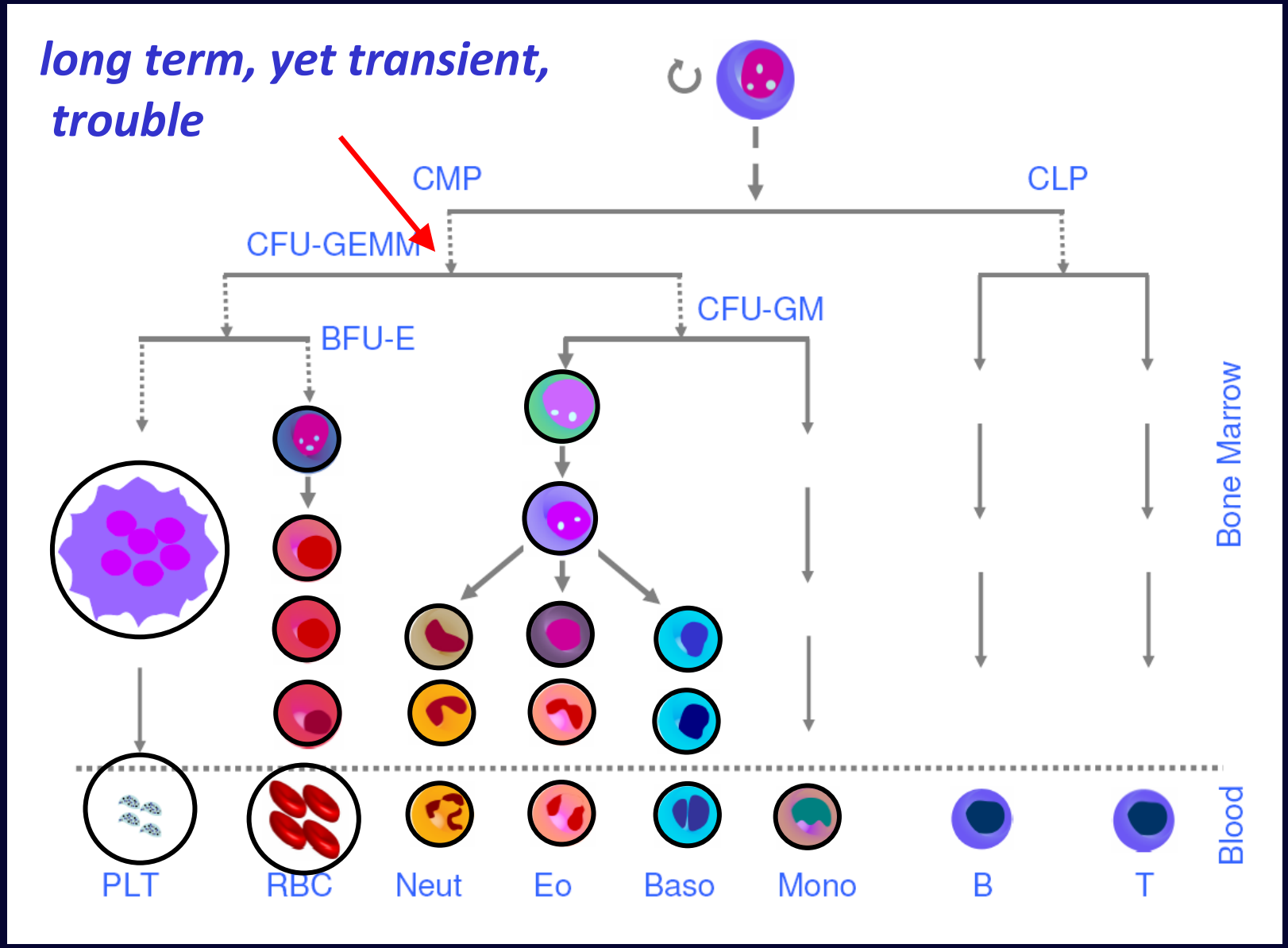


mutations arising at this stage are but ripples in hematopoiesis

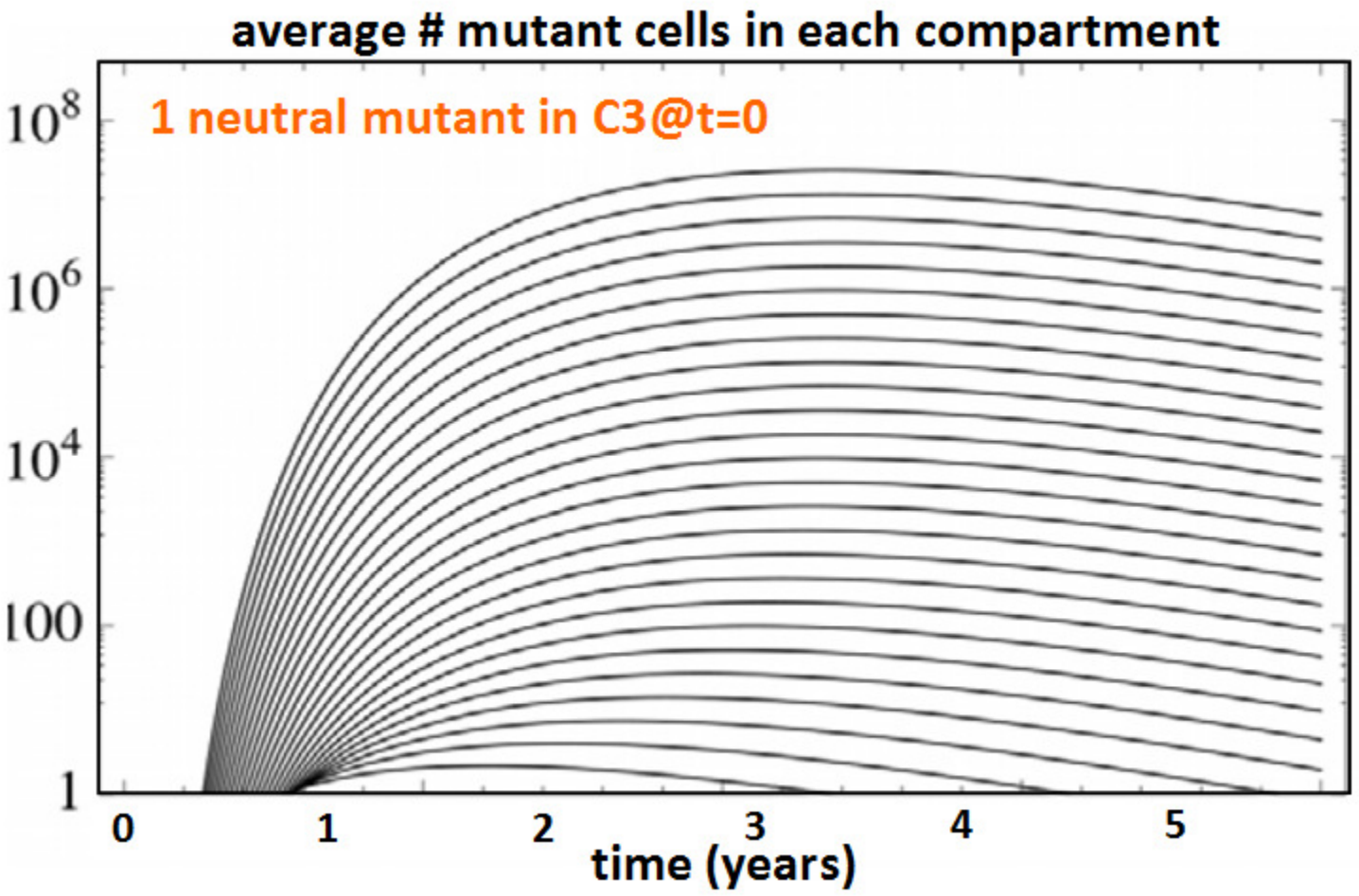
this type of mutations is very likely : up to compartment 14, the total number of cells is already ~10^8

trouble

normal : $10^{-7} < \mu < 10^{-6}$ ***per cell per replication***



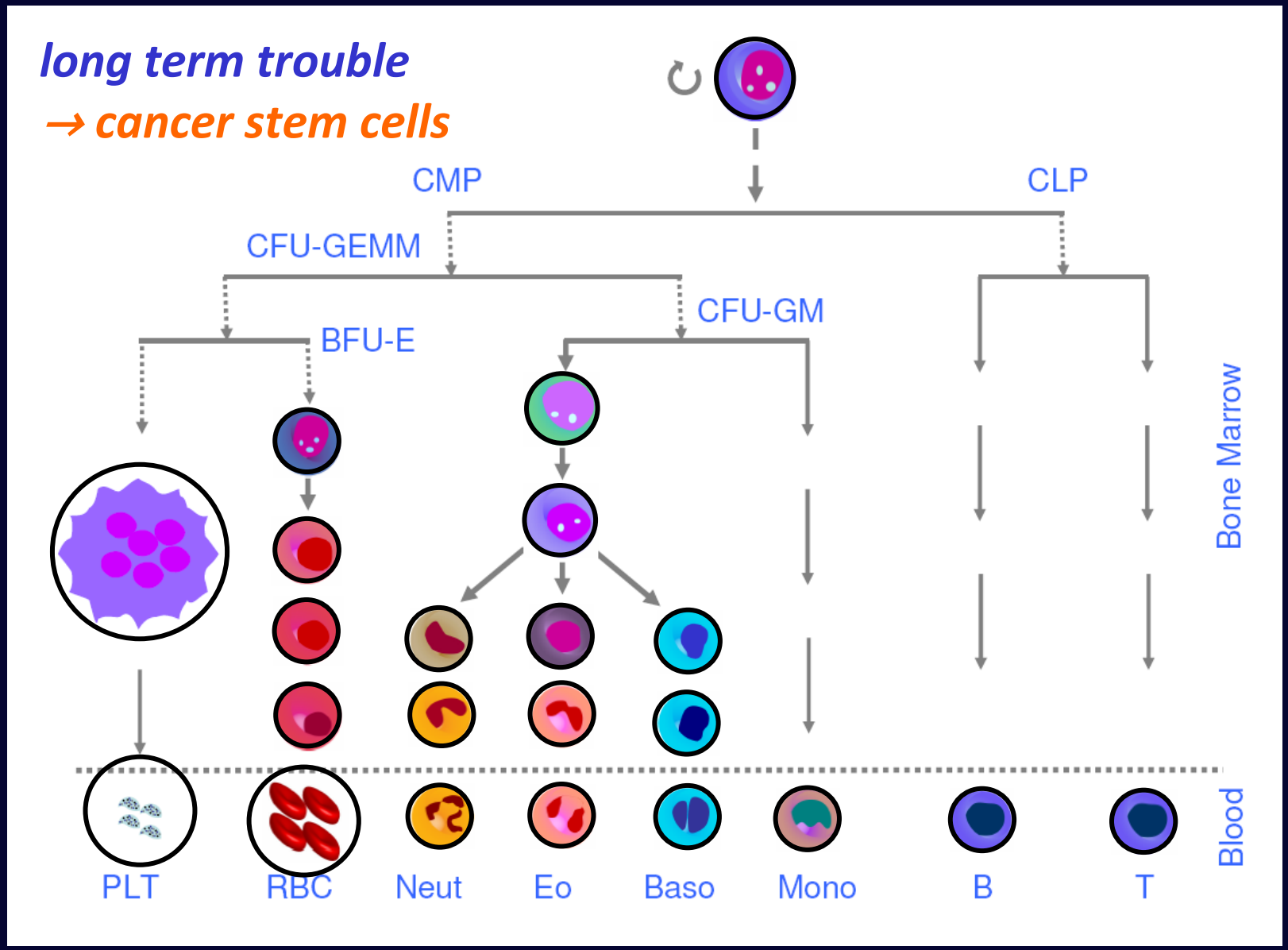
trouble



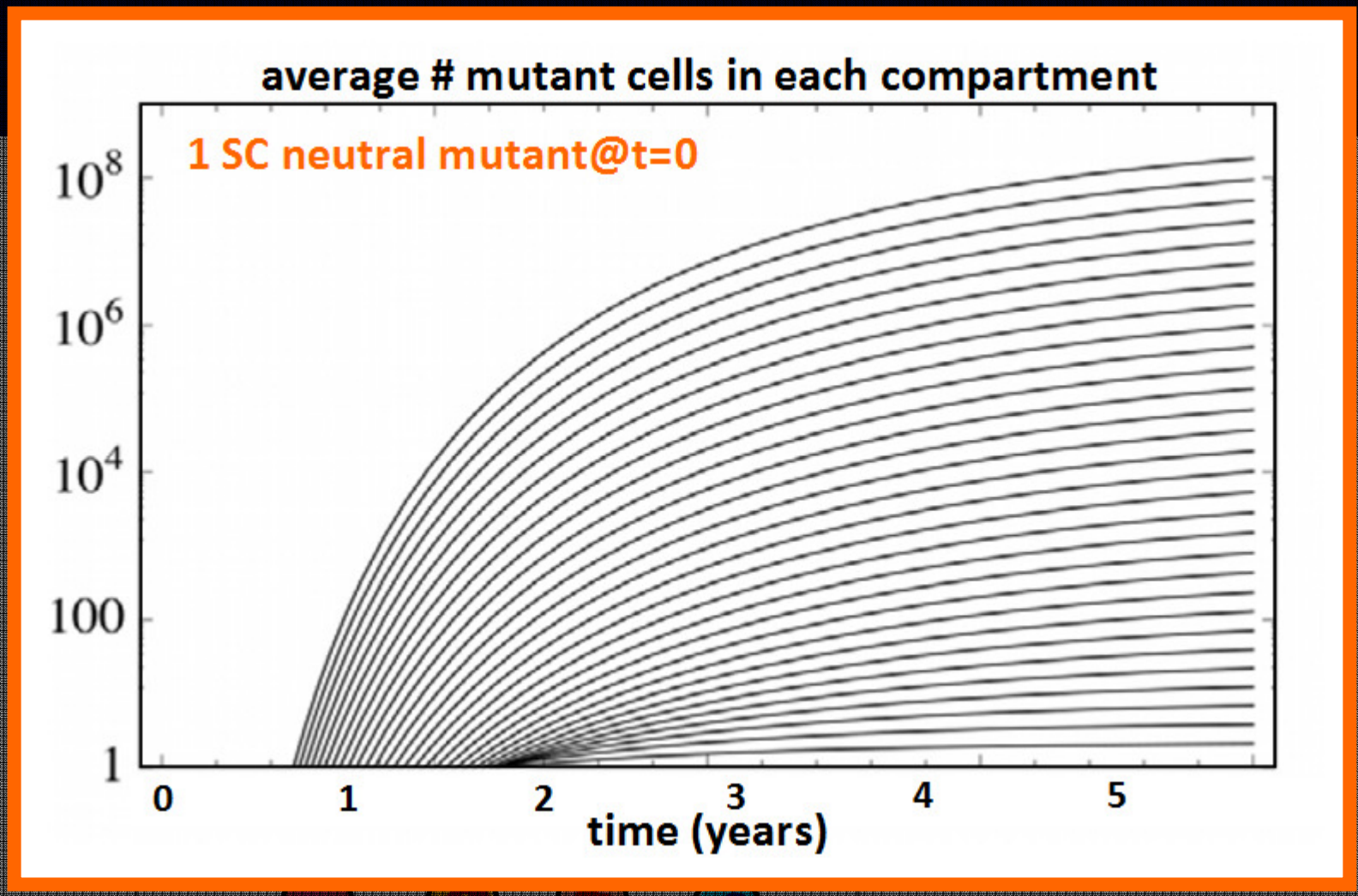
mutations arising in early progenitors may lead to long term trouble but are ultimately washed out (after many years)

trouble

normal : $10^{-7} < \mu < 10^{-6}$ ***per cell per replication***



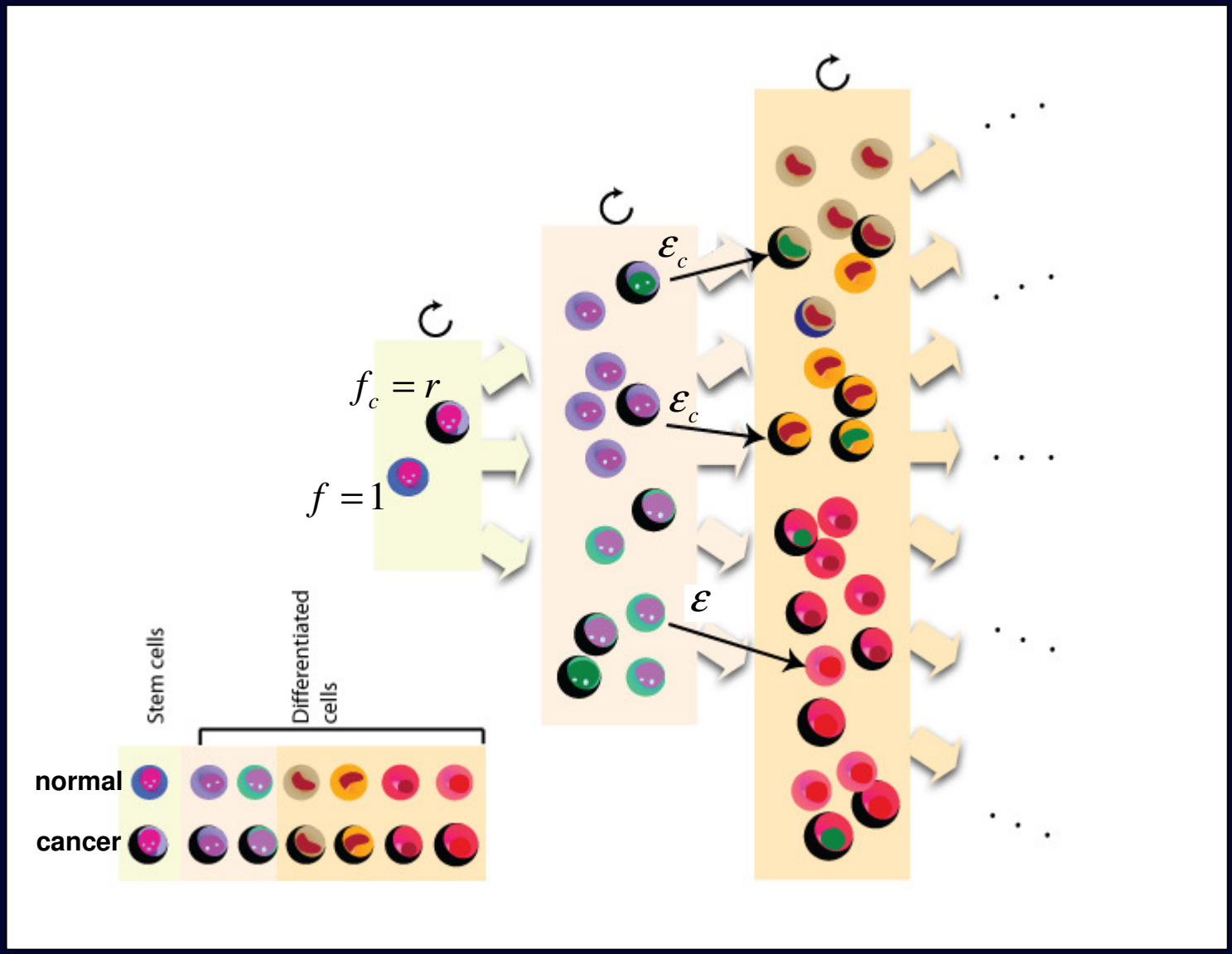
trouble



mutations arising in *HSC* lead to potentially permanent trouble as hematopiesis may evolve toward a new steady state

(stochastic effects may change this)

troubled hematopoiesis

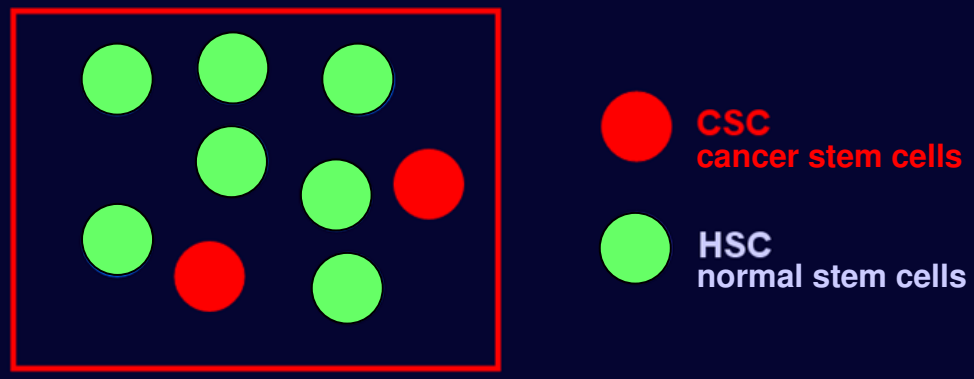


cancer dynamics becomes a multi-scale ecology of cell competition

starting upstream with a small number of HSC & CSC and getting downstream into very large numbers of cells of all kinds

stochastic dynamics of *HSC* (birth-death)

stochastic model for HSC :

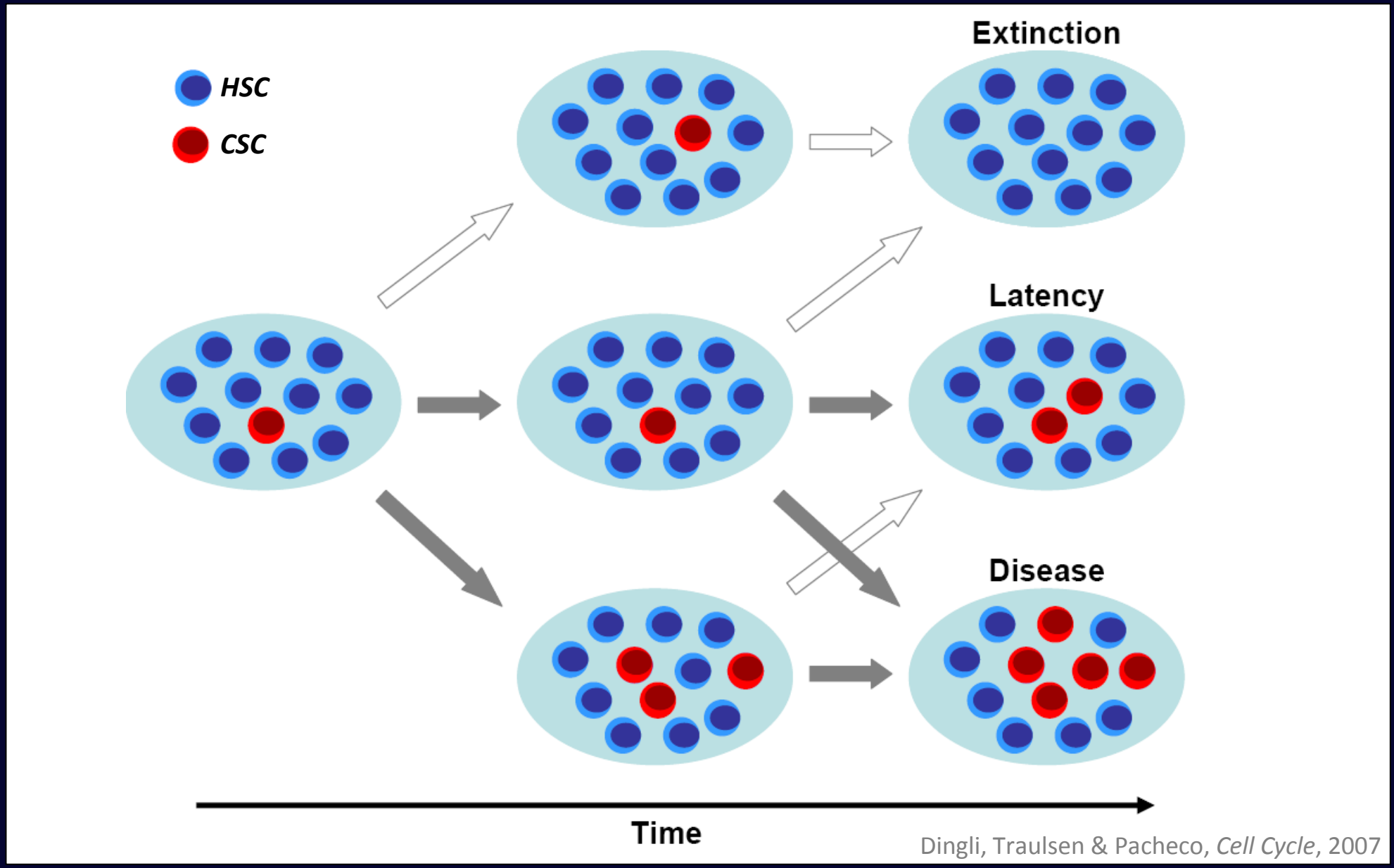


- *SC population remains constant* (16.55 $M^{3/4}$)
- *HSC divide at normal rate* (2.9 $M^{-1/4}$)
- *CSC divide at rate* $r \times$ *normal, where* r *= relative fitness*
- *when a cell is selected , gives rise to two new identical cells*
- *subsequently, 1 cell is randomly selected for export*
- *HSC may suffer mutations and transform into CSC*

Dingli, Traulsen & Pacheco, *Cell Cycle*, 2007

stochastic dynamics of *HSC*

several possible scenarios out of this simple process:



Dingli, Traulsen & Pacheco, *Cell Cycle*, 2007

on the small number of *HSC*

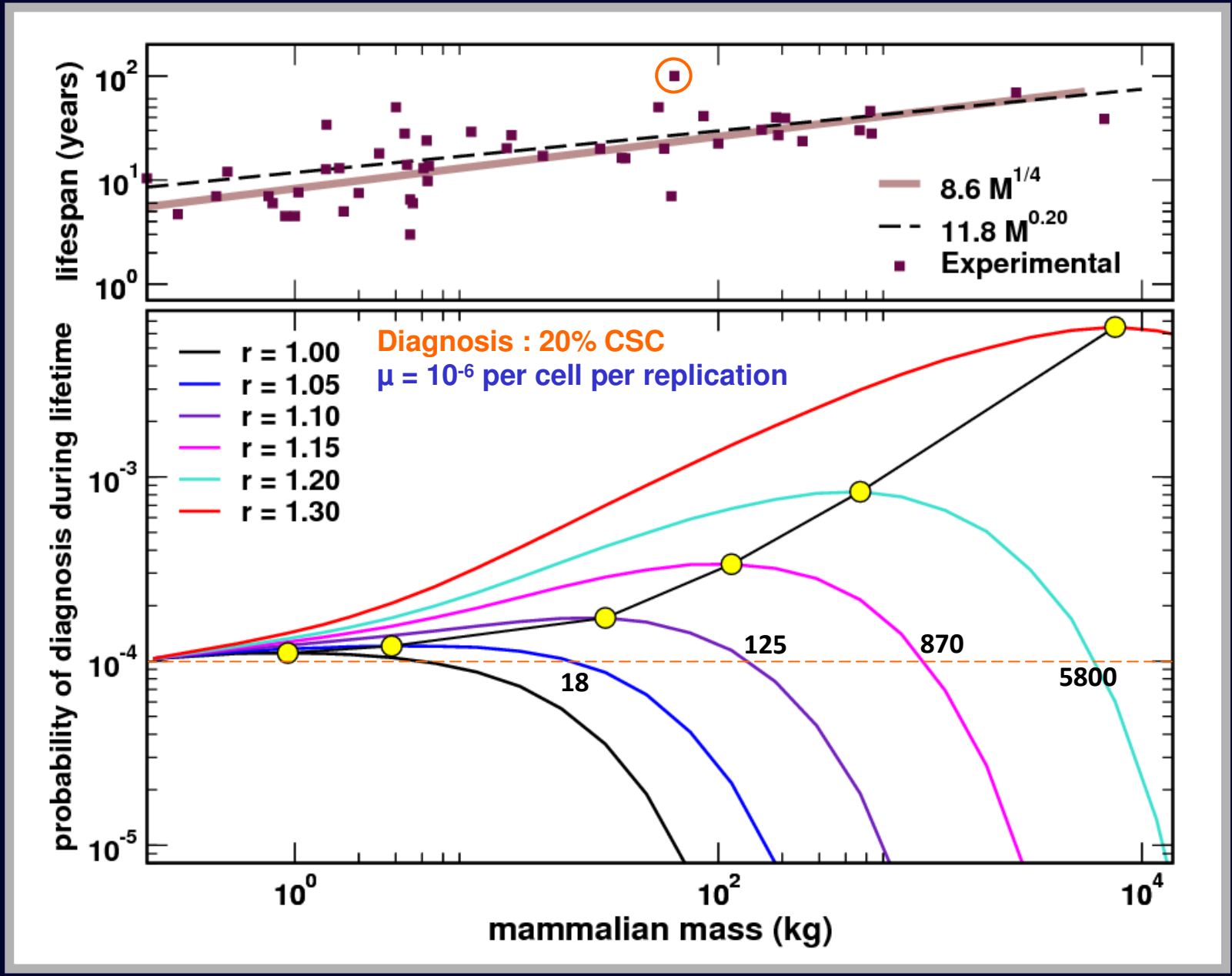
- *a patient is diagnosed in association with some level of disease burden*
- *diagnosis can only happen during the lifetime of the organism*
- *. . . which means there may be no time for diagnosis to happen*
- *for an organism with a finite lifetime, in whch disease means some threshold is surpased, selection & mutation play a curious game . . .*
- *if we assume that disease is equally represented in all cell lineages, we may look at dynamics within the HSC compartment only (not always true)*

the previous model provided average values

stochastic dynamics → time distribution functions
probability during lifetime

on the small number of *HSC*

- *is there a good reason for a small HSC pool ?* (*use scaling for all* **M**)



Lopes, Dingli, & Pacheco, *Blood* 110 (2007) 4120 - 4122

on the small number of *HSC*

- *is there a good reason for a small HSC pool ?*
- *r is usually very difficult to determine experimentally;*
  unfortunately, it is consensual that, in general, r is large (>1.5)
- *when r ~ 1, large mammals are more protected than small mammals;*
- *when r > 1.3, small mammals are more protected, since the probability for the organism to acquire cancer mutations is minimized;*
- *a small active HSC pool minimizes the risk of mutations;*
  once mutations occur, the path to full blown disease opens up easily (whenever r >1).

how about the probability distribution functions ?

Lopes, Dingli, & Pacheco, *Blood* 110 (2007) 4120 - 4122

stochastic dynamics of *HSC* in Humans

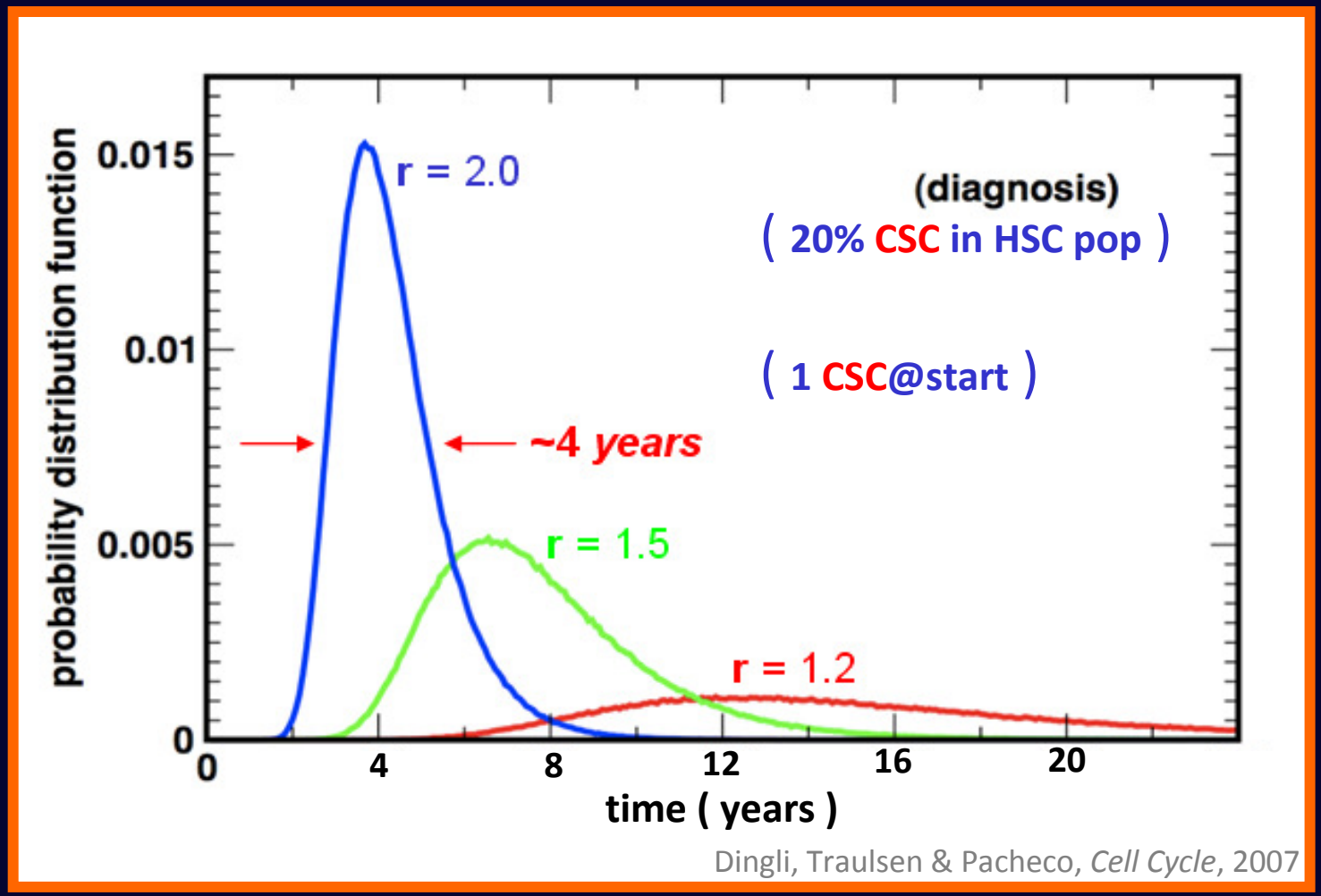
disease diagnosis : ***20% "blasts" in AML***

(acute myeloid leukemia)

10% of plasma cells in MM

(multiple myeloma)

how much time is required for a mutation to develop & give rise to diagnosis of a HSC disorder ?



Dingli, Traulsen & Pacheco, *Cell Cycle*, 2007

stochastic dynamics of *HSC* in Humans

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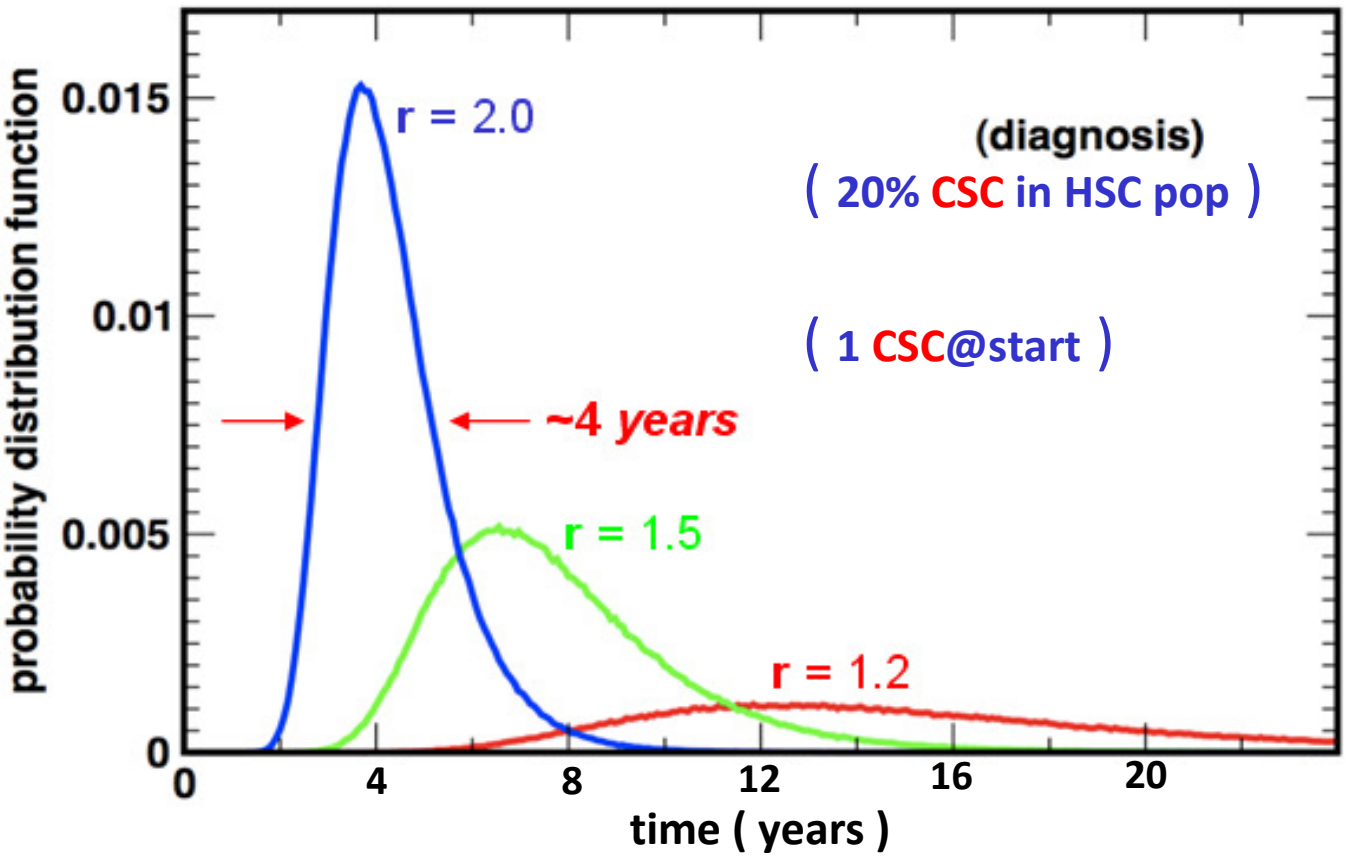
*how much time is required for a **mutation to develop** & give rise to diagnosis of a HSC disorder ?*

even for r=2, the FWHM is ~4 years !



Dingli, Traulsen & Pacheco, *Cell Cycle*, 2007

stochastic effects at play in specific diseases

paroxysmal nocturnal hemoglobinuria

what is known :

- rare disease
- true *HSC* disorder, since it originates in the PIG-A gene of a *HSC*
- rate of PIG-A gene mutation is known to be normal
- often BMF is later observed

conventional wisdom regarding disease development :

- 1st mutation is neutral but a 2nd mutation leads to a fitness advantage of PNH cells → disease expansion (rare event)

Dingli, Pacheco & Traulsen, *Physical Review E77* (2008) 021915

upper limit for the appearance of a 2nd mutation until the first one leads to diagnosis

$$F < \mu N_0 t_M^1 = \mu \frac{N_0^2}{M} \sum_{i=1}^{M-1} \frac{M-i}{N-i} < 10^{-3}$$

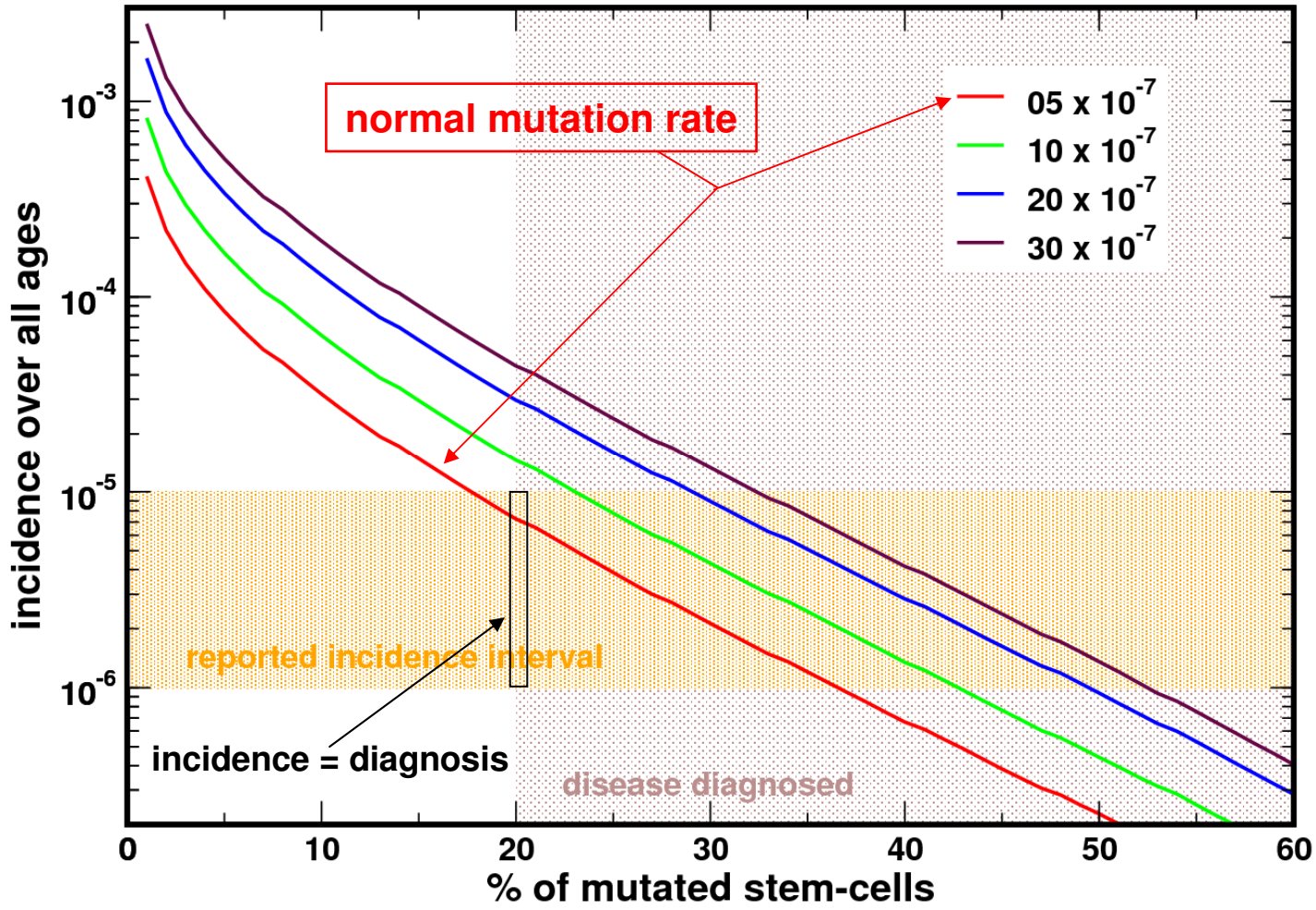
- *relative fitness advantage* of PNH cells due to an imunne attack to normal HSC → disease expansion

PNH - model features

disease development

- use $N_{SC} = 400$
- simulate **HSC** activity in virtual USA (10^9 virtual Americans)
- use normal mutation rate for HSC → PNH transformation
- assume ***neutral drift*** (**r=1**) between **HSC** & **PNH** cells
- fold data with CENSUS 2000 for USA population
- compare results with incidence data in *USA*

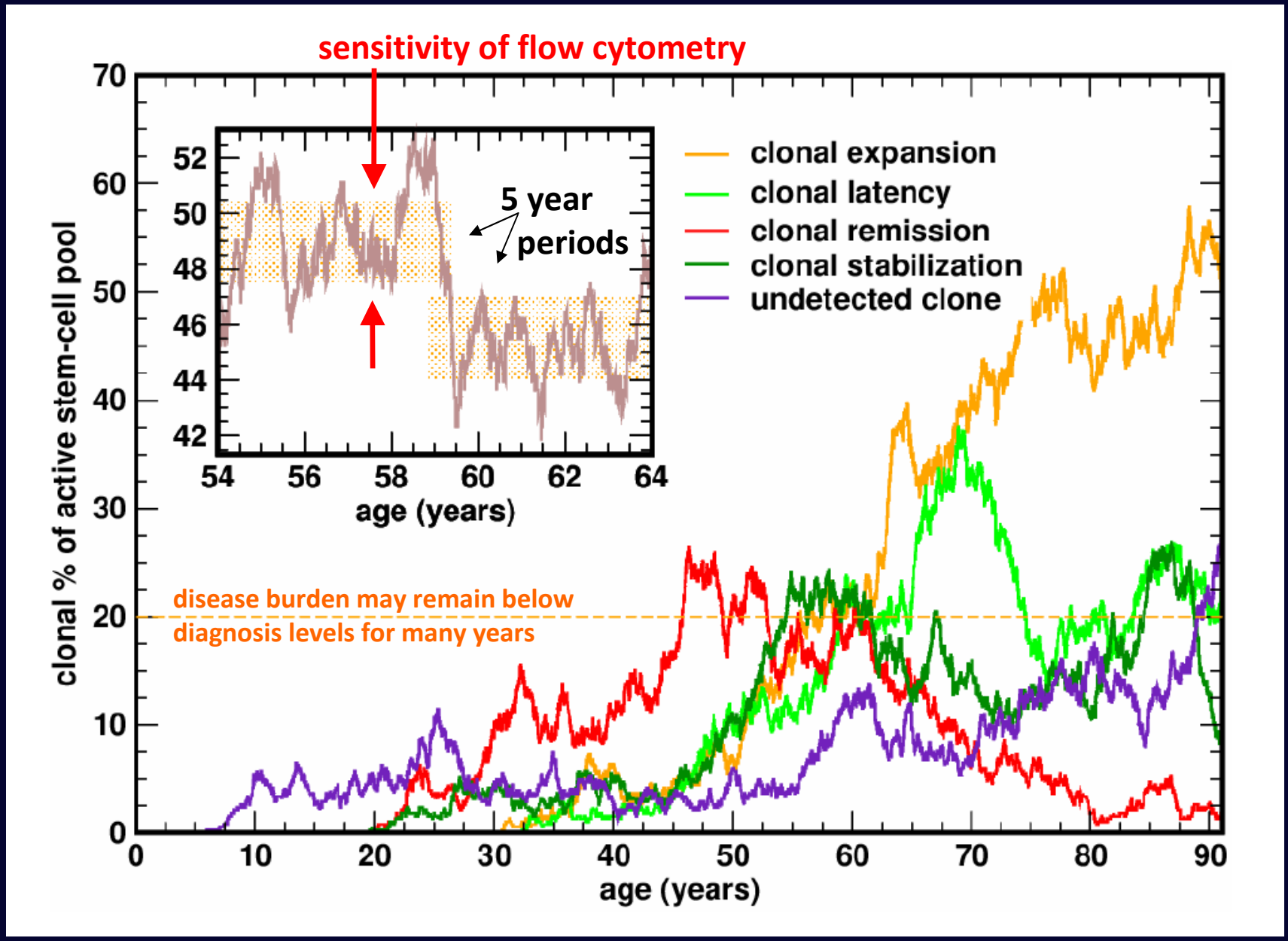
results



results above (& other results) suggest that it is not necessary to invoke a relative fitness difference to explain incidence of PNH

Dingli et al, *PNAS* 2008

results – individual history & variable outcomes



Dingli et al, *PNAS* 2008

neutral evolution relies on the stochastic nature of cell behavior, & **PNH** shows us that, likely, many individuals suffer the PIG-A mutation but are never diagnosed PNH, as it is more likely for the mutant to become extinct than to evolve into a clone. This, in turn, suggests that the current way of approaching the (now over) 40-year old war-on-cancer, that is,

cure = ***kill-every-single-cancer-cell***

is perhaps not alwayss the best, or sometimes maybe even unnecessary.
In fact, profiting from competition through natural selection may turn out to be a more viable strategy.

Chronic Myeloid Leukemia

what is known :

- Hematopoietic stem cell disorder
- Initial event: Philadelphia chromosome
- ? HSC are enough to drive chronic phase ?
- clonal expansion and myeloproliferation
- stem cell derived but progenitor cell driven
- *abl*-kinase inhibitors very effective

CML dynamics

- **Q-RT-PCR data from patients treated with *imatinib***
- **2 data sets available**
  - Michor *et al, Nature*, 2005
  - Roeder *et al, Nature Medicine*, 2006
  - other data recently available for ***nilotinib***
- **data fitting (using deterministic model)**

CML - model features

disease development

- use existing model of hematopoiesis
- how to get from HSC origin to progenitor driven disease ?
- bone marrow expansion $\longrightarrow \varepsilon_{CML} < \varepsilon_0$

treatment

- how does *imatinib/nilotinib* work ?
- does *imatinib/nilotinib* induce cell death?
- how many cells are responding to *imatinib/nilotinib* ?

CML - model constraints

disease development

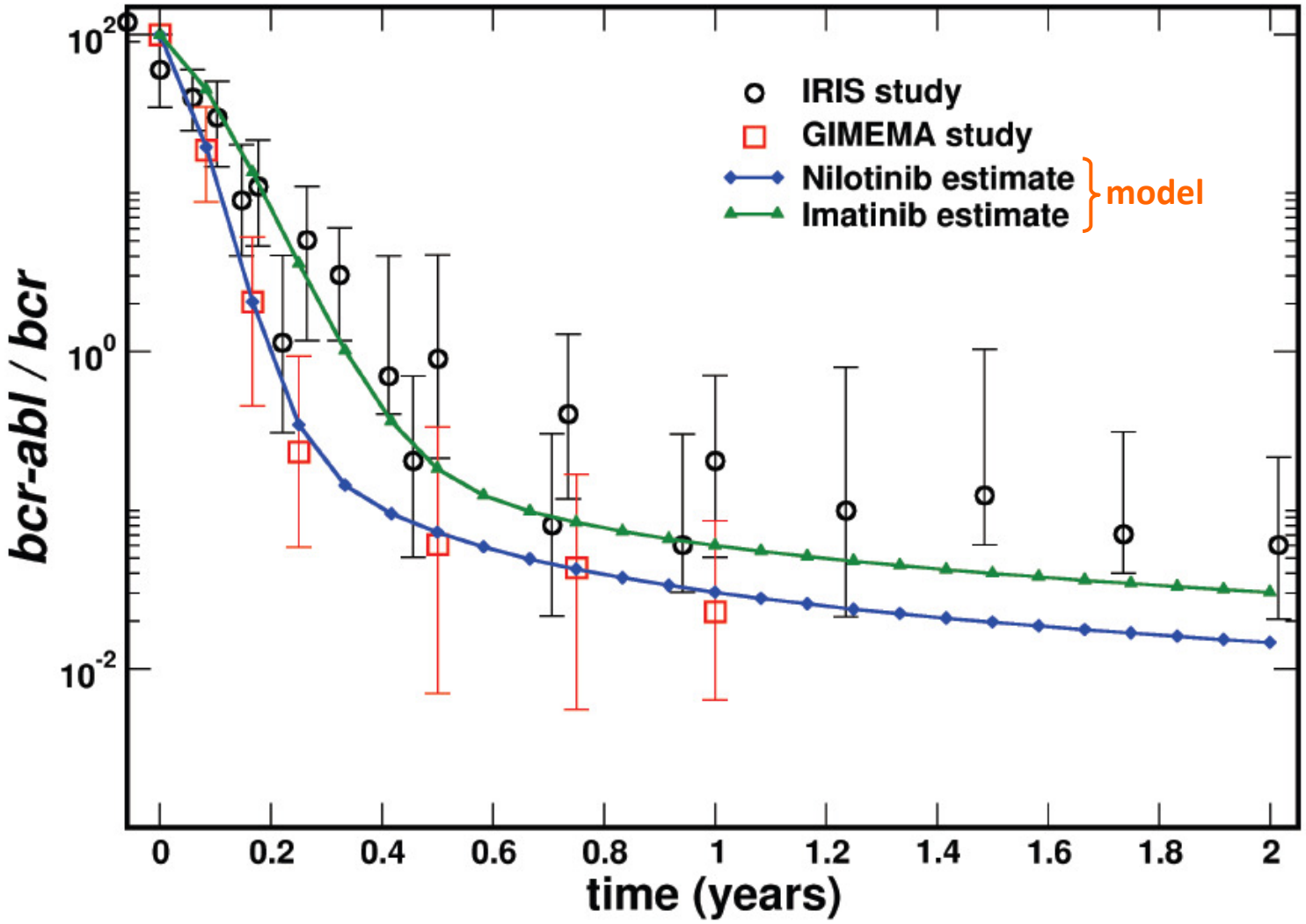
- time from initial insult to diagnosis is 3.5 – 6 years
- progenitor cell expansion >14%
- total number of active *HSC* is *not* increased
- daily bone marrow output is ~ 3 x normal

treatment

- *imatinib/nilotinib* leads to $\varepsilon_{IMAT} > \varepsilon_0 > \varepsilon_{CML}$
- *imatinib/nilotinib* does not affect HSC
- at any time a fraction *z* of cells responds to *imatinib/nilotinib*

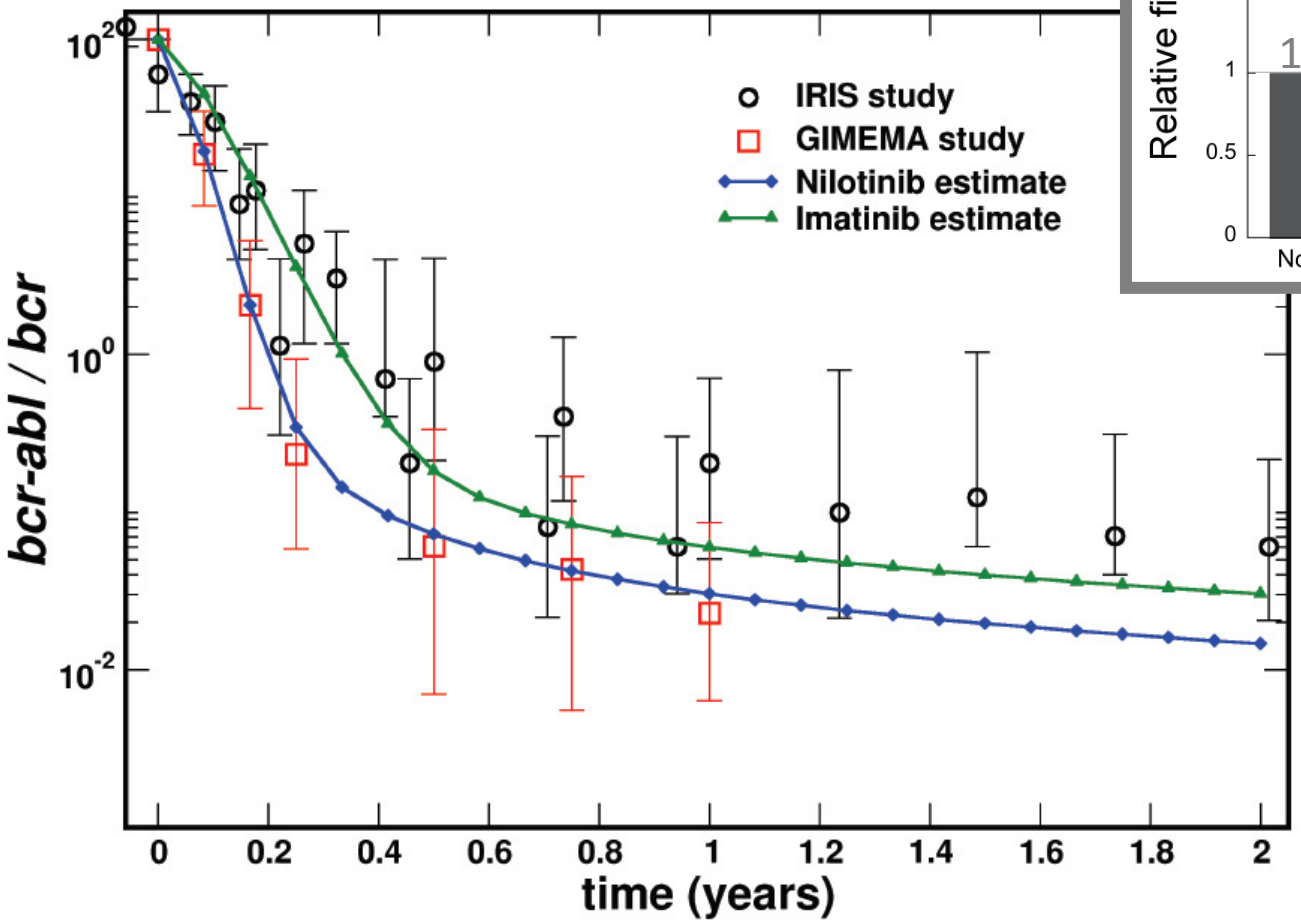
we extract $\varepsilon_{IMAT} > \varepsilon_0 > \varepsilon_{CML}$ & *z* from data …

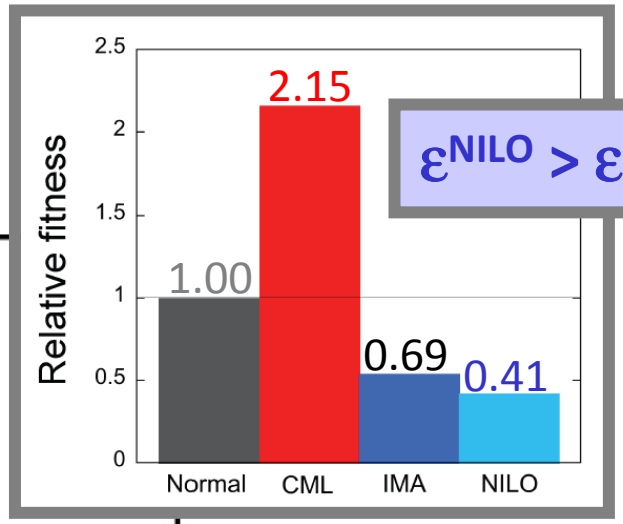
imatinib ⊗ nilotinib



Traulsen *et al, Cancer Letters*, 2010

imatinib ⊗ nilotinib





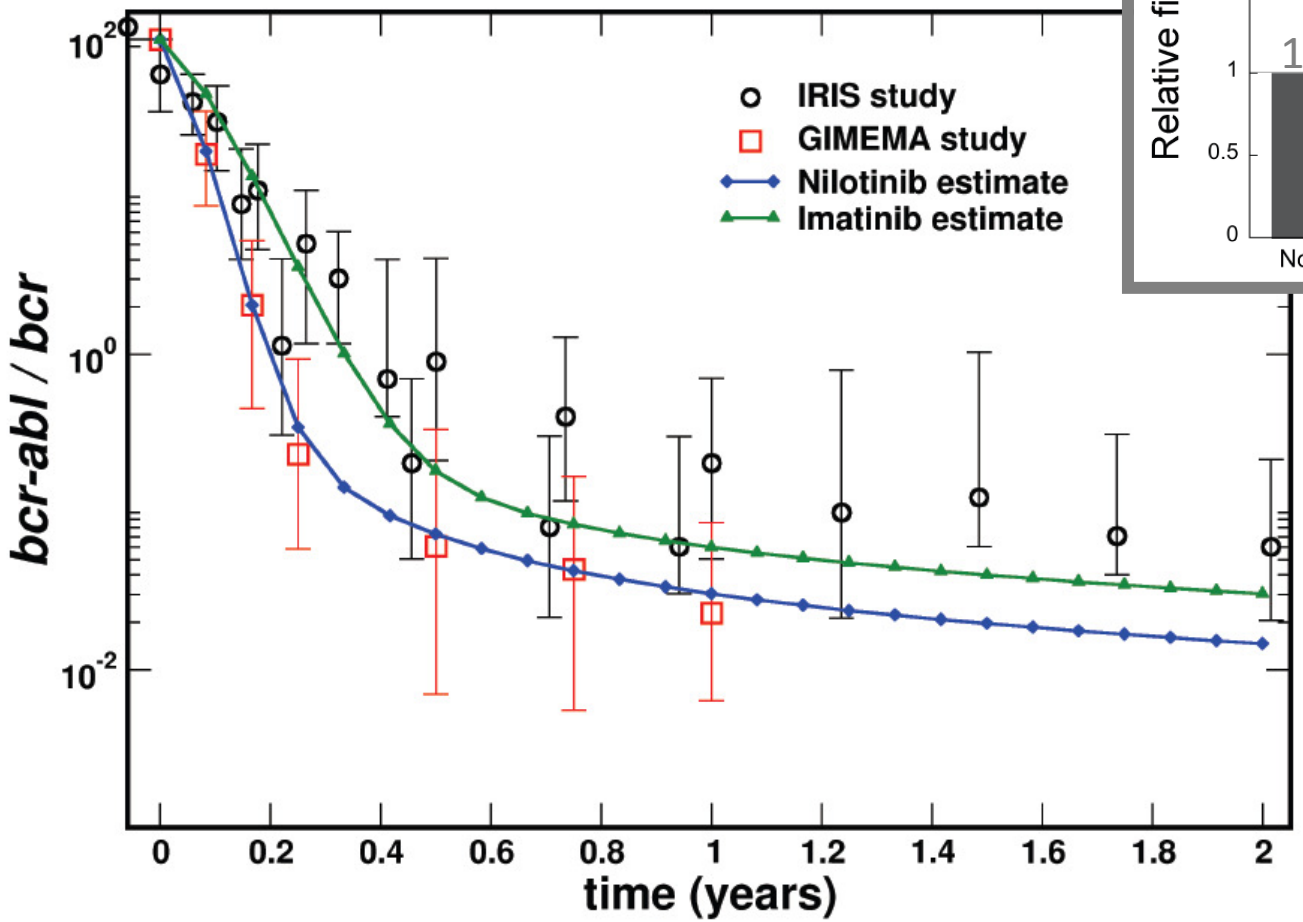
$\varepsilon^{NILO} > \varepsilon^{IMAT}$

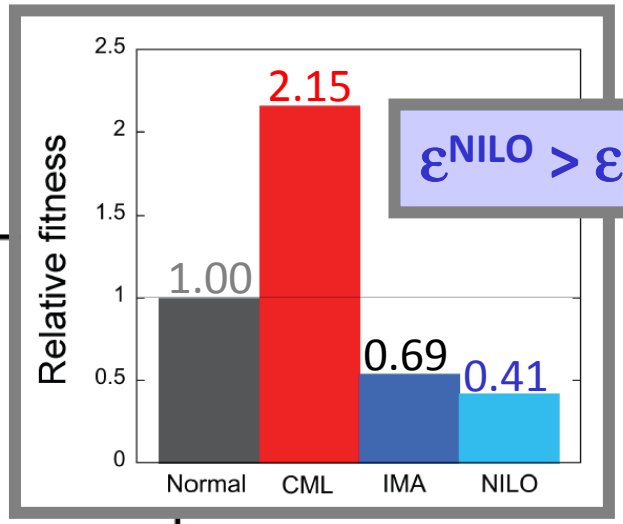
$$f_j = \frac{\rho_j}{\rho_0} = \frac{1-\varepsilon_j}{1-\varepsilon_0}\frac{\varepsilon_0}{\varepsilon_j}$$

Traulsen *et al, Cancer Letters*, 2010

imatinib ⊗ nilotinib

in vitro **: *no* differences detected**
in vivo **: *important* differences**
(ecology of cancer cells is important)





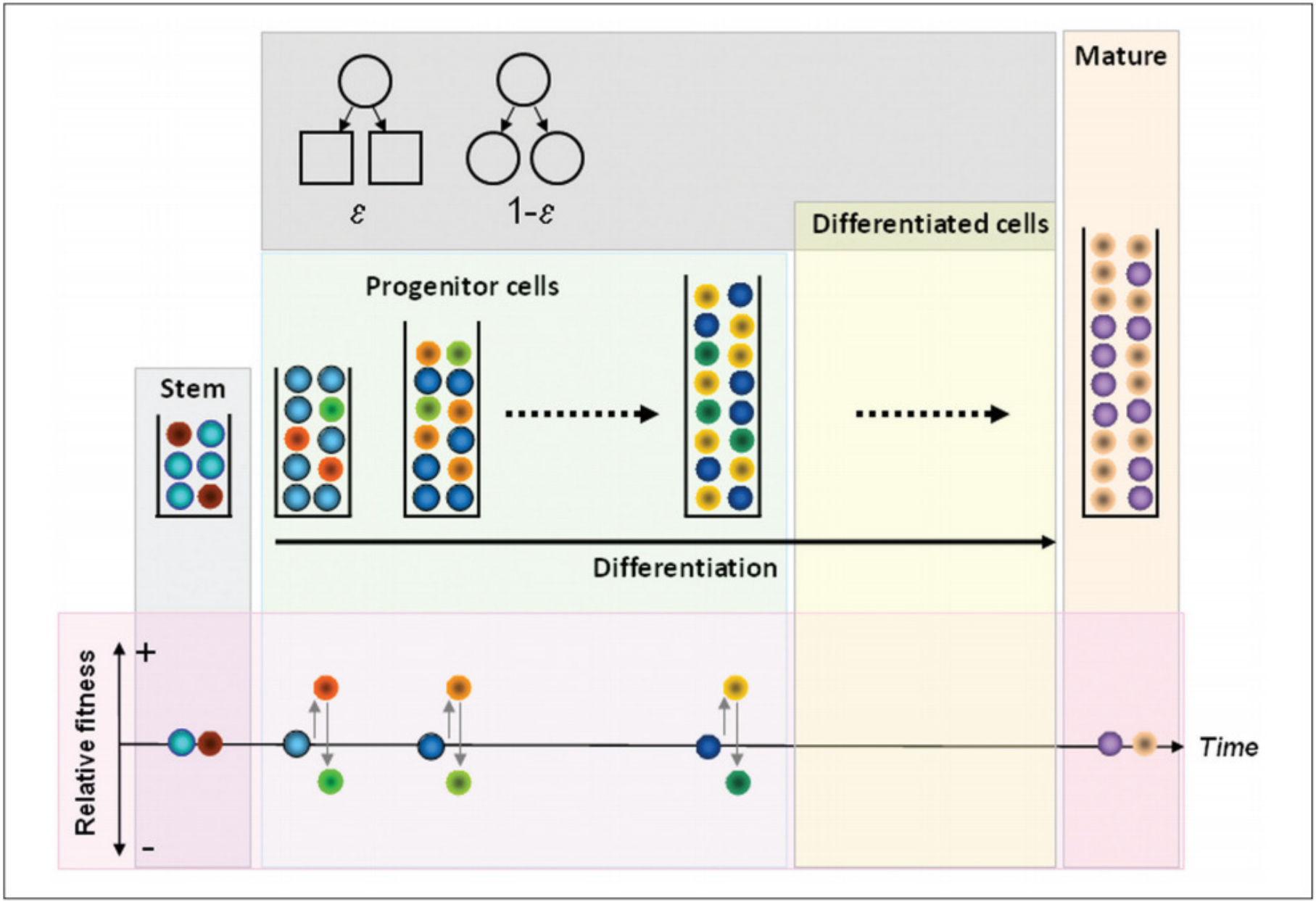
$\varepsilon^{NILO} > \varepsilon^{IMAT}$

$$f_j = \frac{\rho_j}{\rho_0} = \frac{1-\varepsilon_j}{1-\varepsilon_0}\frac{\varepsilon_0}{\varepsilon_j}$$

neat way to deduce fitness from data

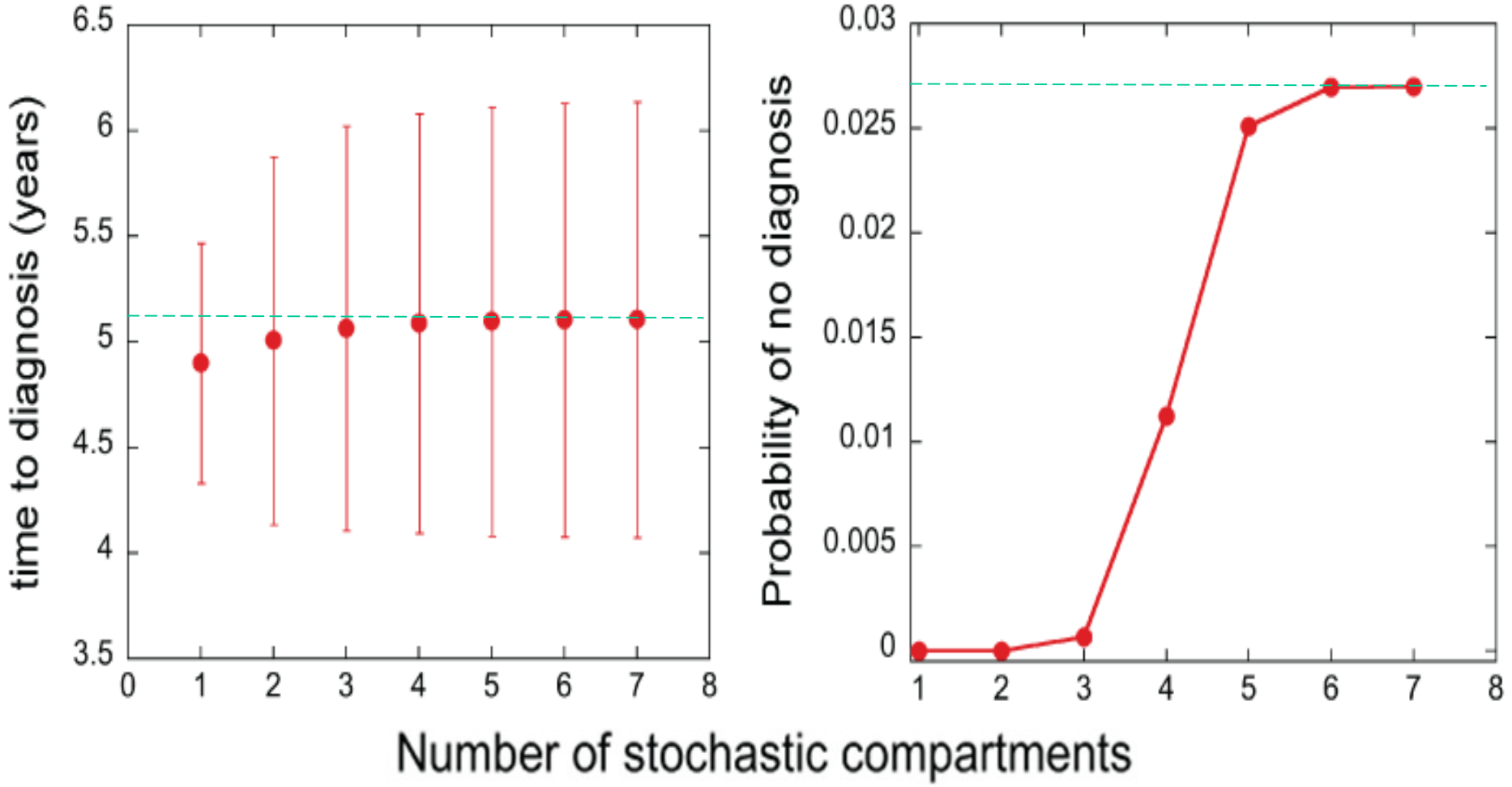
Traulsen *et al, Cancer Letters*, 2010

evolutionary dynamics of CML



stochasticity in CML

. . . stochastic dynamics of 10^{12} cells is unfeasible → ***hybrid model***

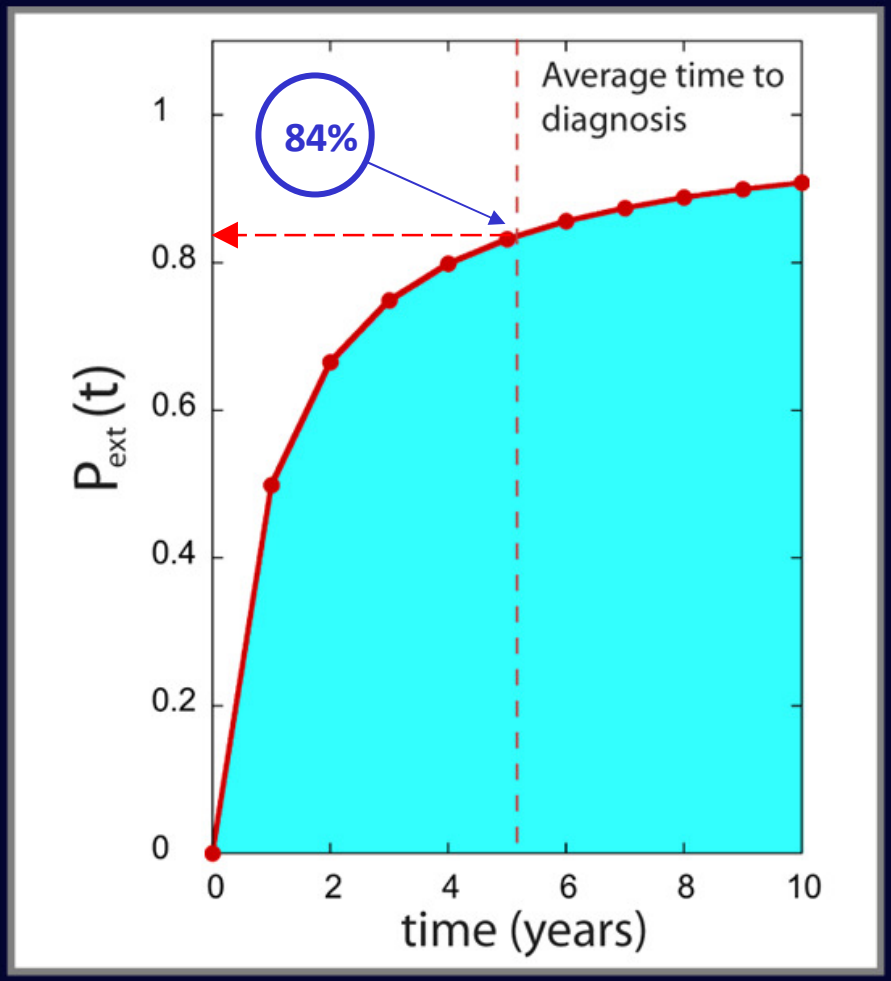


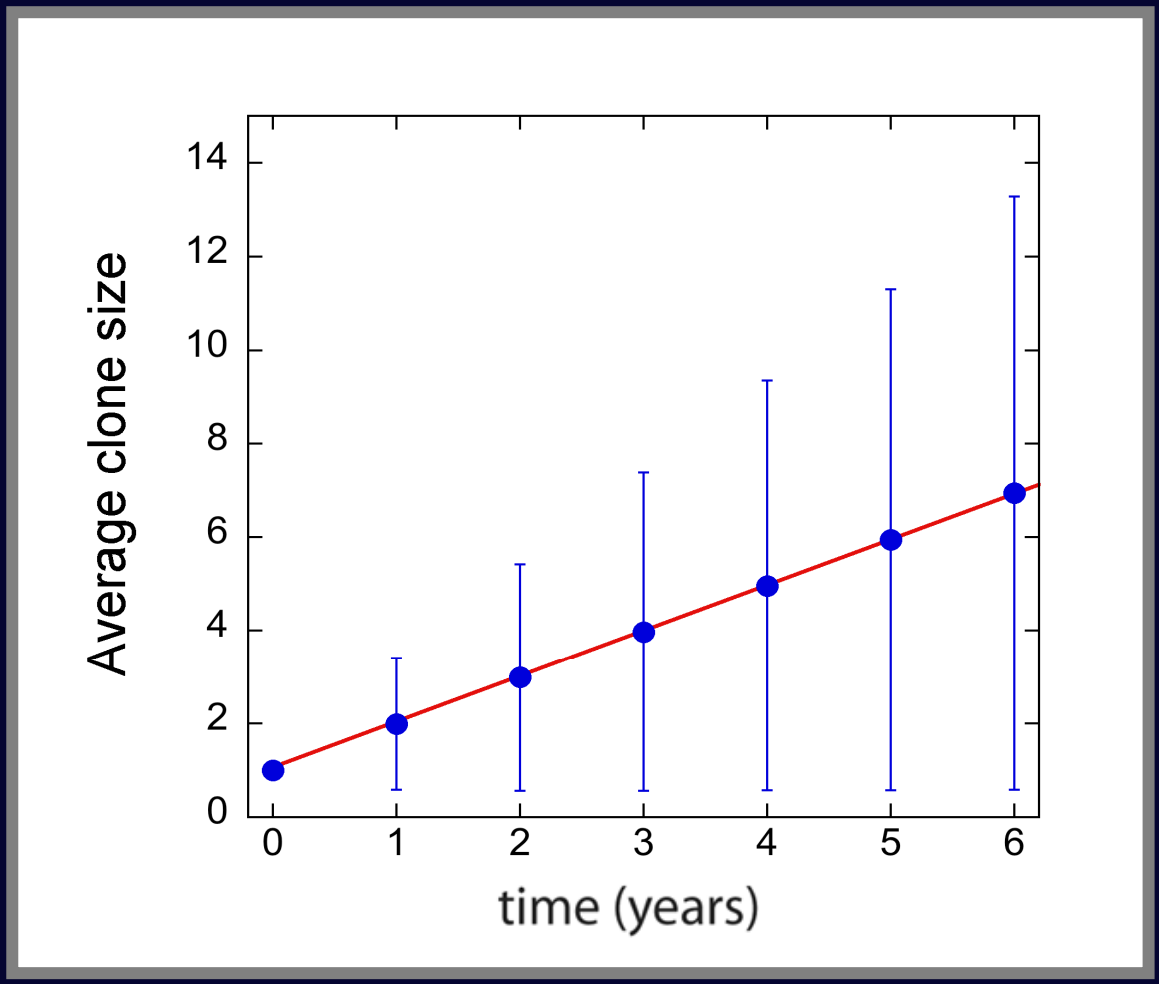
Tom Lenaerts *et al.*, *Haematologica* 2010

stochasticity in CML

in 84% of individuals, CSC population goes extint before diagnosis

in 16% of individuals, CSC population grows, on average, 1 per year





Tom Lenaerts *et al.*, *Haematologica* 2010

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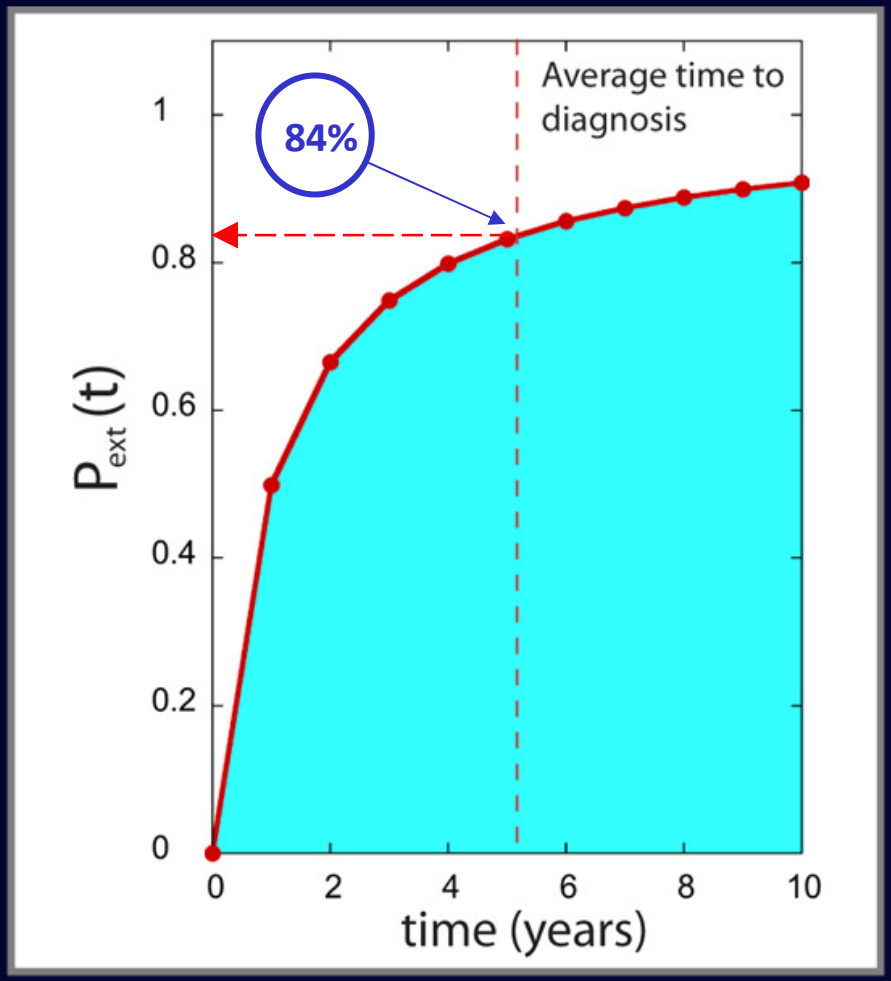
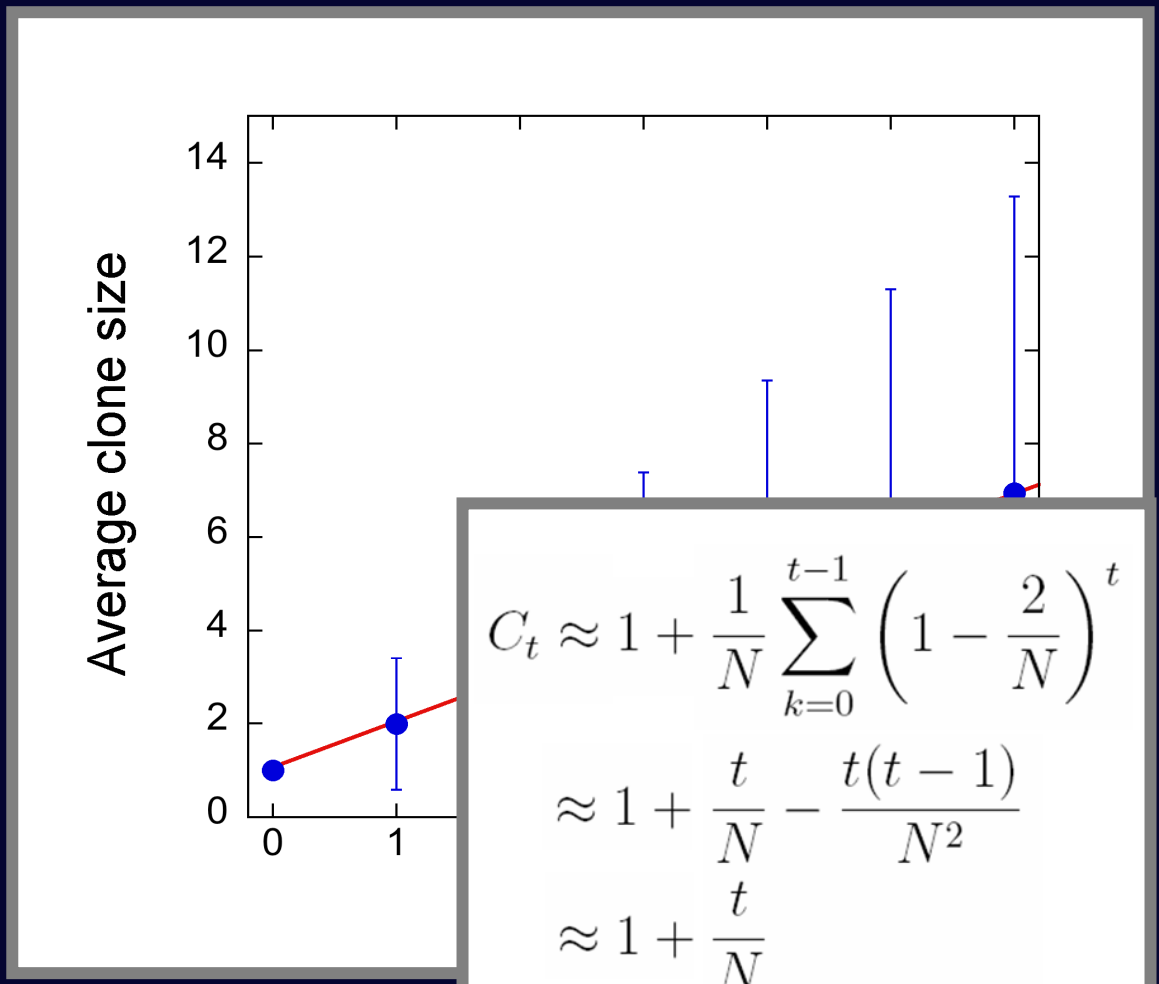




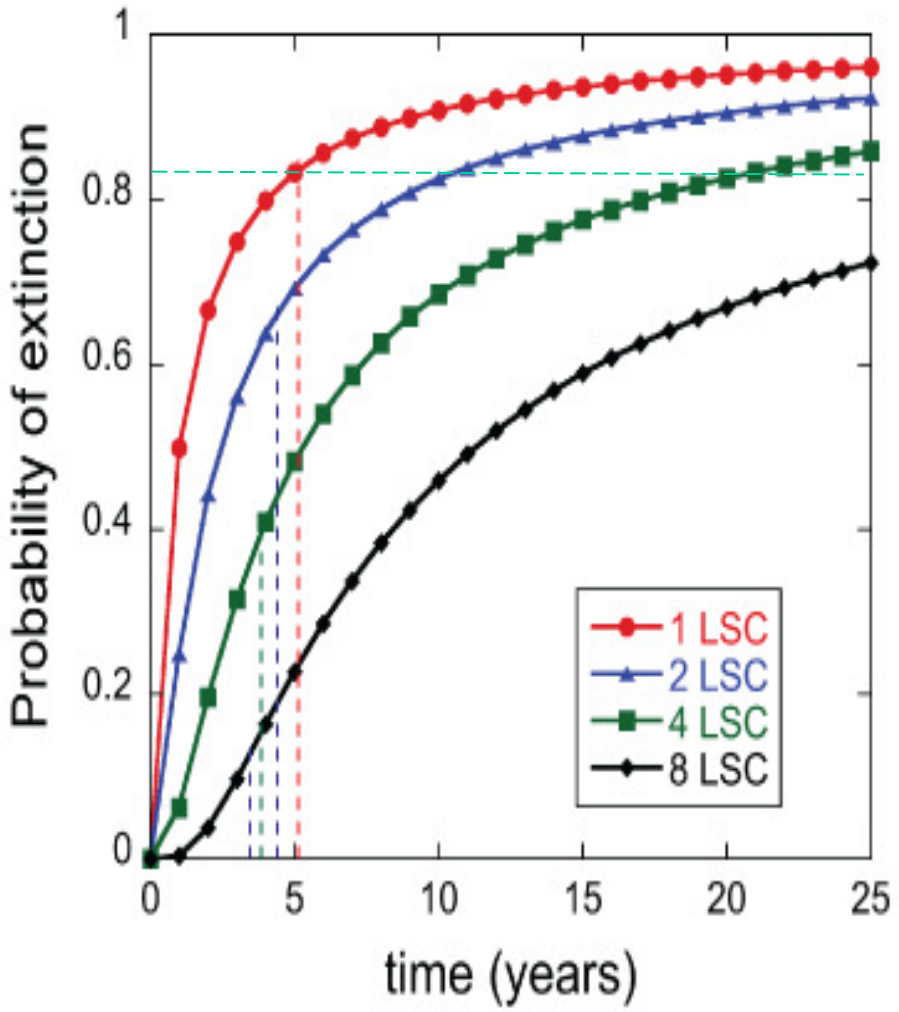
$$C_t \approx 1 + \frac{1}{N}\sum_{k=0}^{t-1}\left(1-\frac{2}{N}\right)^t$$

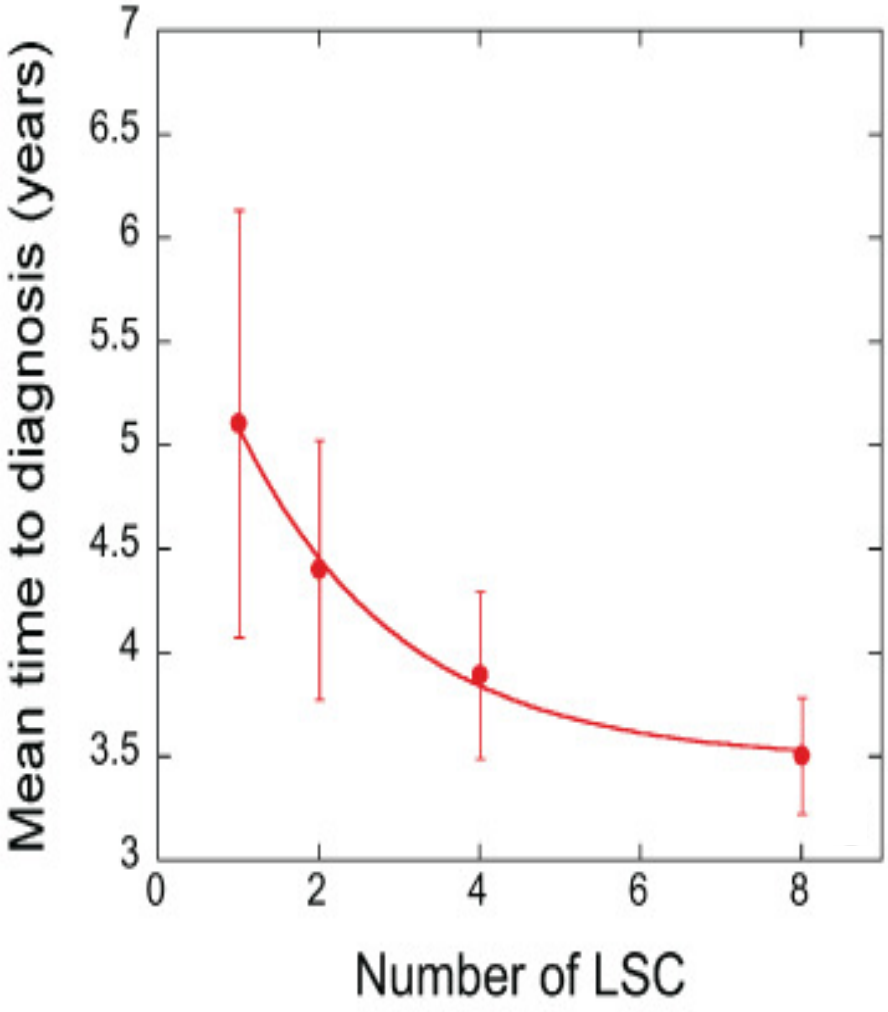
$$\approx 1 + \frac{t}{N} - \frac{t(t-1)}{N^2}$$

$$\approx 1 + \frac{t}{N}$$

Tom Lenaerts *et al.*, *Haematologica* 2010

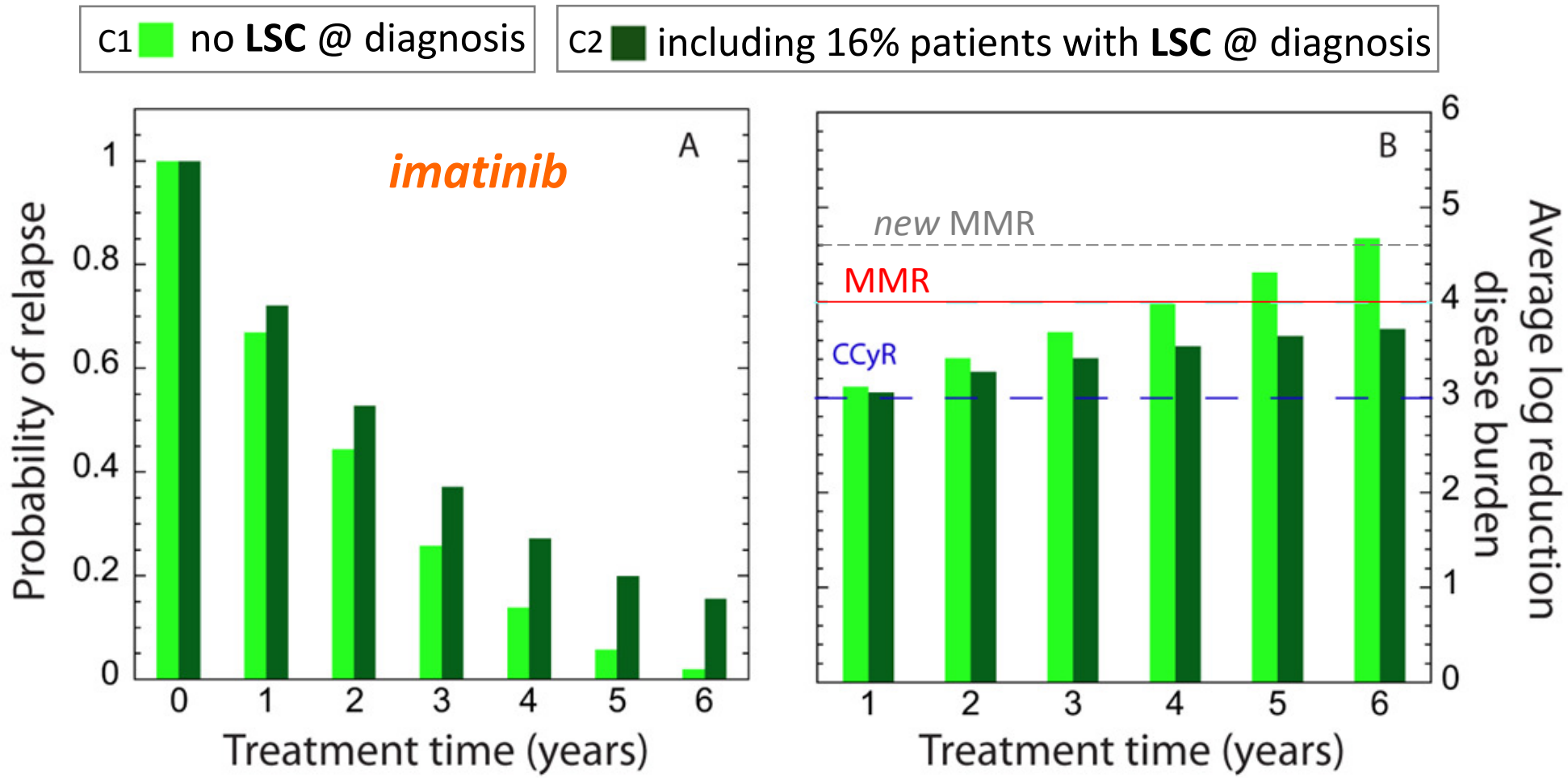
stochasticity in CML





Tom Lenaerts *et al.*, *Haematologica* 2010

stochasticity in CML



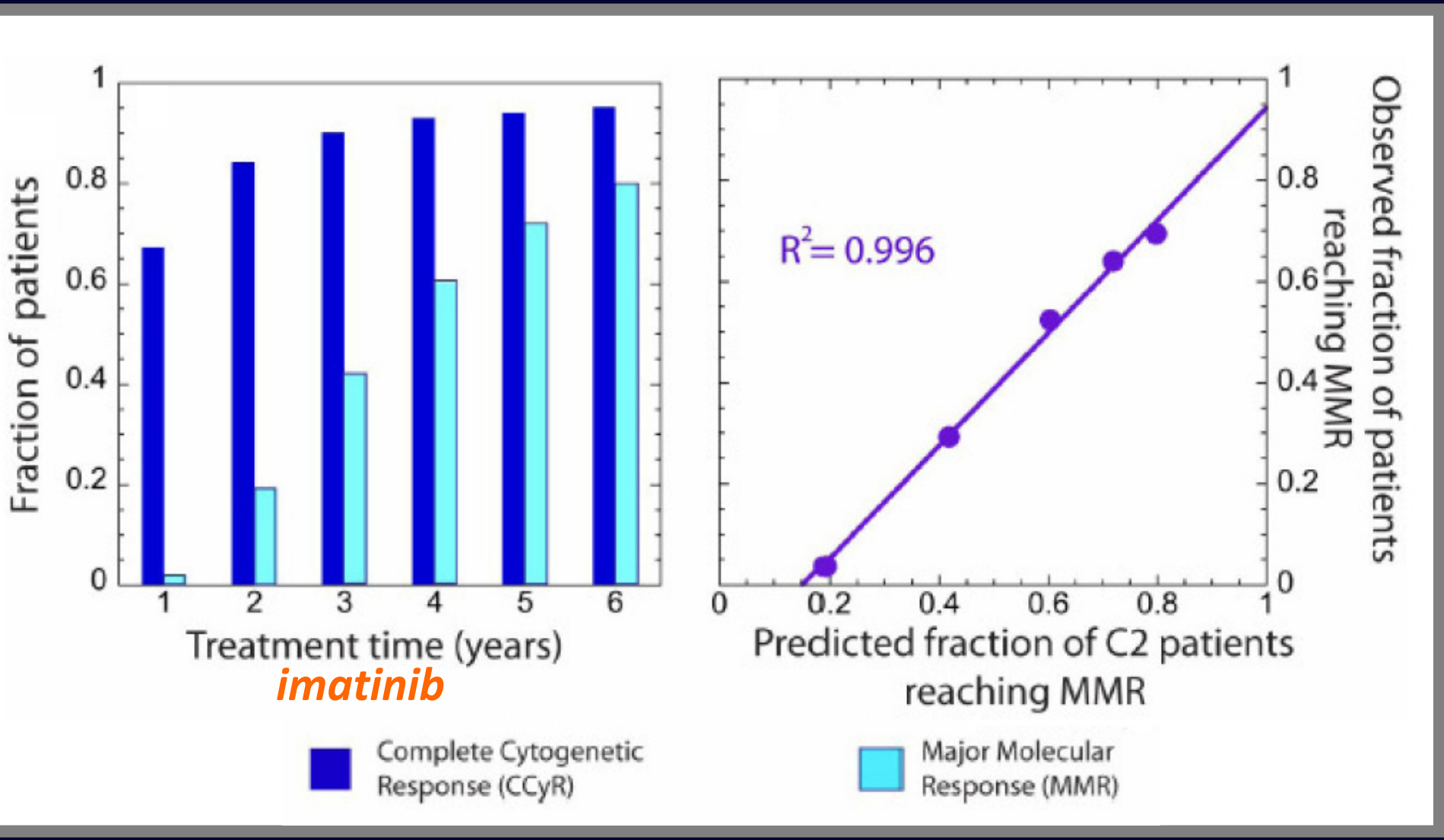
despite *NOT* affecting directly CSC,

imatinib + natural selection can cure the majority of CML patients

ongoing: development of *resistance mutations* . . .

stochasticity in CML

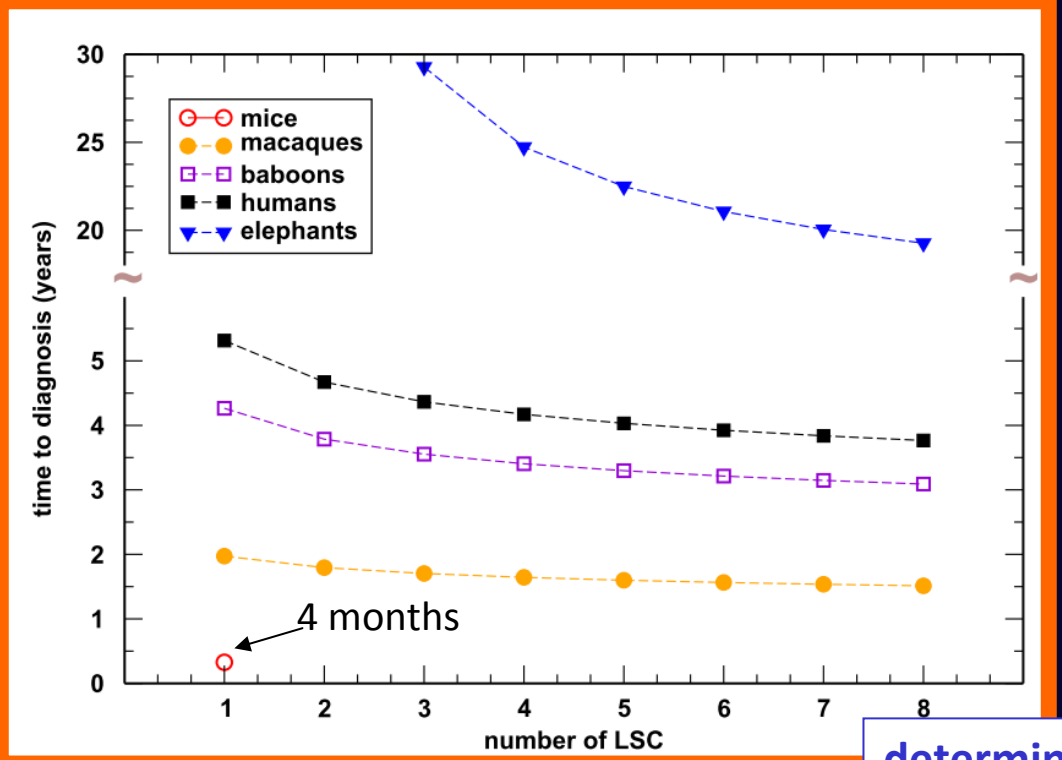
treatment with TKI-inhibitors helps an individual to stay alive and live his everyday life while natural selection helps him getting rid of the cause of the disease; however, it takes years for one to gamble his way out of cancer.

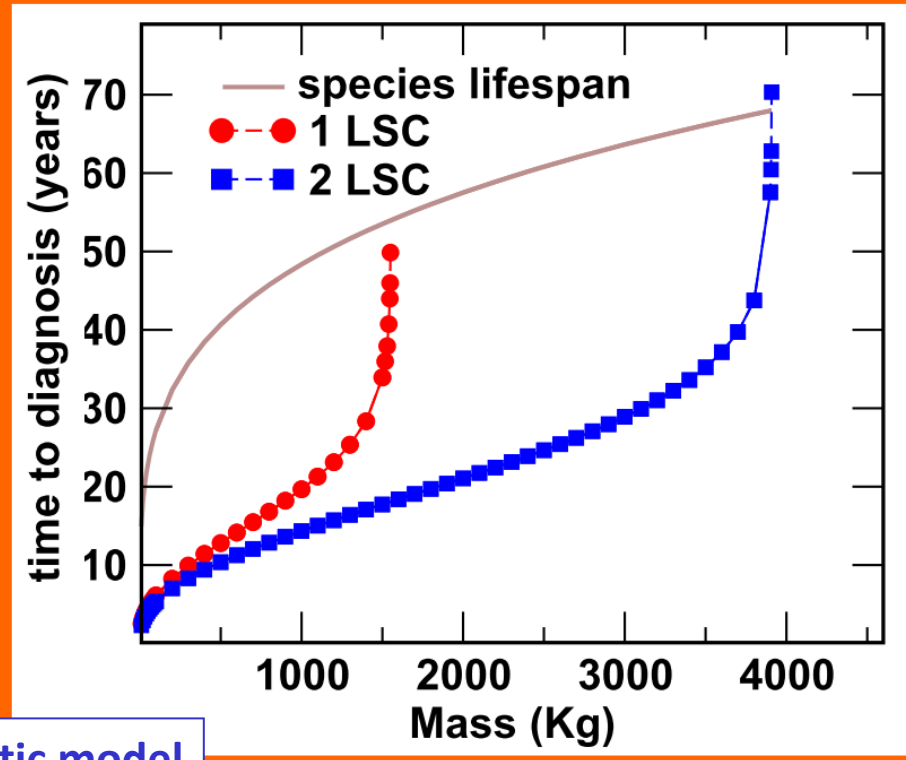


CML in other mammals

there is no reason, *a priori*, to suppose that what we observe in humans stays with humans; how will CML proceed in other mammals ?

- ***HSC* population remains constant (16.55 $M^{3/4}$)**
- ***HSC* & *CSC* divide at normal rate (2.9 $M^{-1/4}$)**
- ***how many CSC drive (or are required to drive) CML in other mammals ?***
- ***how many compartments will behave stochastically in other mammals ?***

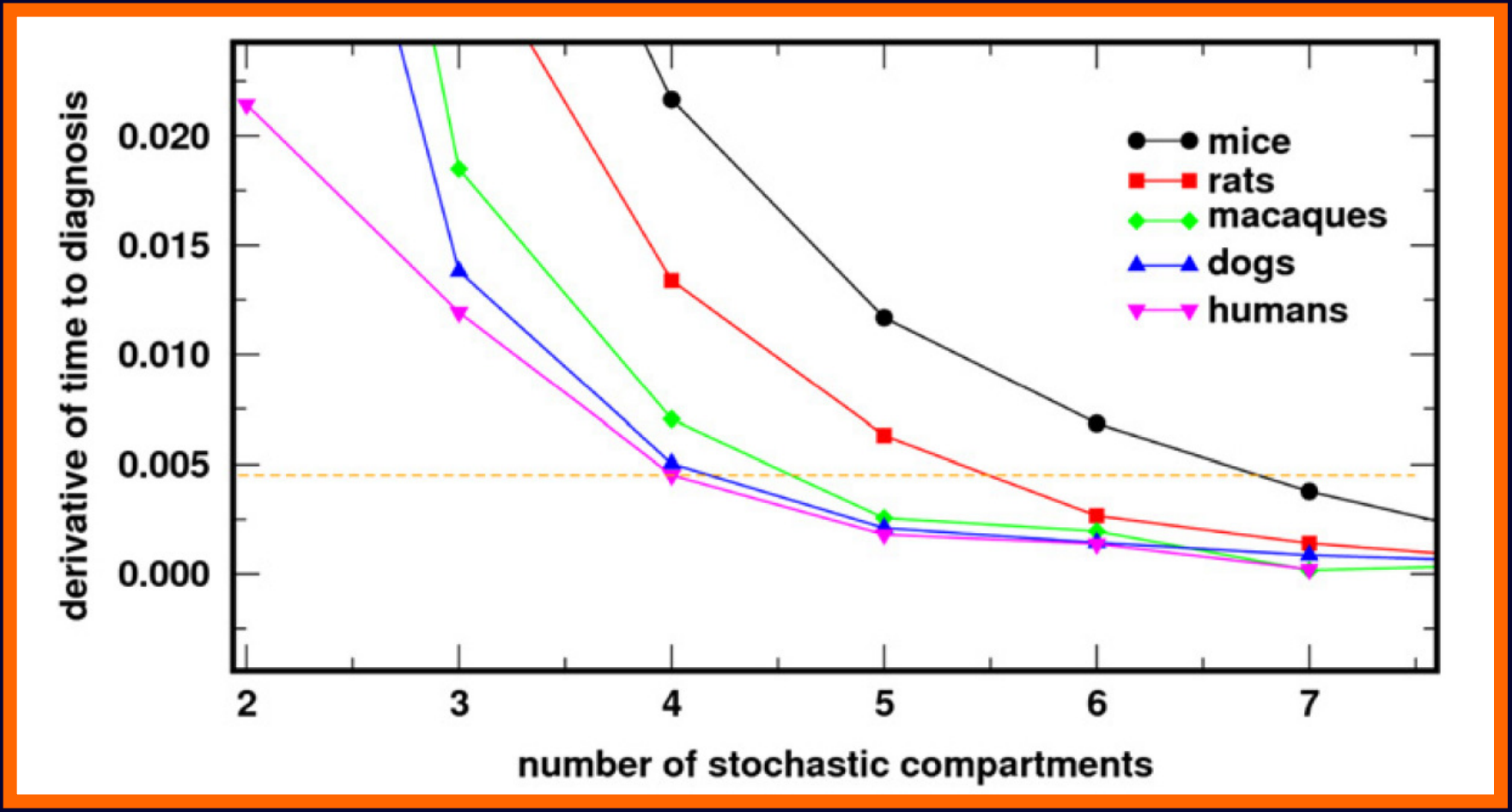




deterministic model

CML in other mammals

to which extent do stochastic effects remain important in CML on other mammals ?



forward difference formula for the derivative of the TTD as a function of the # of compartments treated stochastically, taking as reference K=4 for humans & 1 CSC@start.

conclusions

- *across mammals, hematopoiesis is generated by numbers of **HSC** that may change significantly from species to species, flowing downstream in a multi-compartmental tree-structure in which consecutive compartments interact*

- *in this simple model, homeostasis is nothing but the stationary solution of the coupled problem.*

- *this coupled dynamics, together with specific thresholds for disease diagnosis and the finite lifespan of organisms leads to a complex interplay between selection and mutation in hematopoiesis . . .*

- *. . . where stochastic effects may play an important role and, in some cases, a crucial one.*

- *in some rare **HSC** diseases (ex: CML), evolutionary dynamics of the disease may favor the patient to get rid of its cause, but this alone may not be enough & treatment may be crucial to keep patient alive*

E N D