

Mutational Robustness

A mini-review with applications

Luca Peliti

Dipartimento di Scienze Fisiche and Sezione INFN
Università di Napoli “Federico II”



Natural Selection

Darwin, 1859

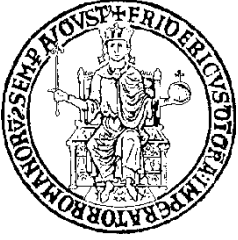
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Natural Selection

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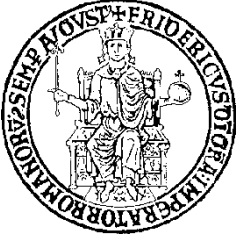
This preservation of favourable variations and the rejection of injurious variations, I call Natural Selection. Variations neither useful nor injurious would not be affected by natural selection, and would be left a fluctuating element, as perhaps we see in the species called polymorphic.



Outline

Mutational robustness : The capacity of a mutated genome to be as fit as the original one

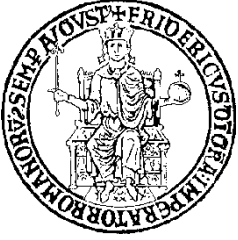
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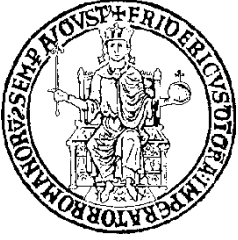
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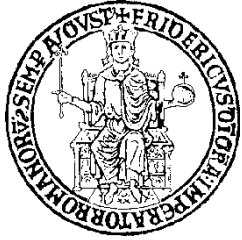
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- ⑥ Sampling and "drift" : Neutral and almost neutral evolution



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- ⑥ Sampling and "drift" : Neutral and almost neutral evolution
- ⑥ Application : Evolution of transcription factor binding sites

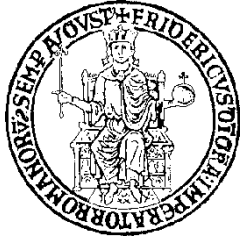


Wright-Fisher sampling

Population of M^0 individuals, with N_G genotypes.

W_g : *fitness* of genotype g

Genotype distribution in the new population ?



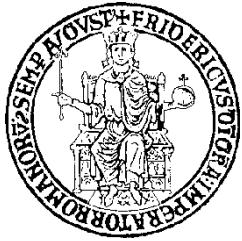
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Wright-Fisher sampling

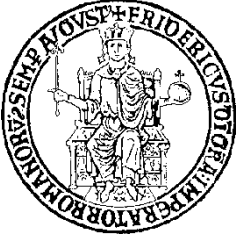
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Genotype distribution in the new population ?

Hypotheses :

- ⑥ Haploid population with one-parent reproduction mechanism
- ⑥ Parent and offspring have the same genotype (i.e., there are no **mutations**)
- ⑥ Reproduction events are independent from one another



Distributions

n_g^0 : number of individuals with genotype g in the old population

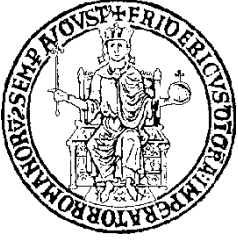
W_g : fitness of genotype g

Expected number of individuals with genotype g in the new generation :

$$\nu_g = N n_g^0 W_g / \sum_{g'} n_{g'}^0 W_{g'}$$

Probability that the number of individuals with genotype g in the new generation is equal to n_g :

$$P_g(n_g) = \frac{e^{-\nu_g}}{n_g!} \nu_g^{n_g}$$



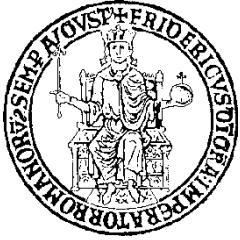
Fisher's Fundamental Theorem

Fisher, 1925

Hypothesis : $\nu_g \gg 1, \forall g \Rightarrow n_g \simeq \nu_g \propto n_g^0 W_g = N x_g^0 W_g$

$$x_g := \frac{n_g}{\sum_{g'} n_{g'}} \simeq \frac{x_g^0 W_g}{\sum_{g'} x_{g'}^0 W_{g'}}$$

$$\begin{aligned} \langle W \rangle &:= \frac{\sum_g n_g W_g}{\sum_{g'} n_{g'}} = \sum_g x_g W_g \\ &= \frac{\sum_g x_g^0 W_g^2}{\sum_{g'} x_{g'}^0 W_{g'}} = \frac{\langle W^2 \rangle_0}{\langle W \rangle_0} \geq \langle W \rangle_0 \end{aligned}$$



Escaping from Fisher's Theorem

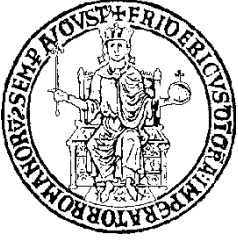
Fisher's hypotheses cannot be held :



Escaping from Fisher's Theorem

Fisher's hypotheses cannot be held :

- ⑥ Mutation rate does not vanish \Rightarrow Quasispecies Theory
- ⑥ Finite population size \Rightarrow Kimura-Ohta theory



Escaping from Fisher's Theorem

Fisher's hypotheses cannot be held :

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What effects they have on the adaptation level ?



Elements of Quasispecies theory

Eigen, 1971

- ⑥ Problem : How do mutations qualify Fisher's adaptation ?
- ⑥ Hypotheses : Nonvanishing (and even large) mutation rate μ , very large population size (deterministic limit)
- ⑥ The process takes place in **sequence space** : sequences of length L with symbols belonging to a \mathcal{A} -letter alphabet (usually $\mathcal{A} = 4$, sometimes $\mathcal{A} = 2$)

$$\Sigma := \{g\} = \{A, C, G, T\}^L$$



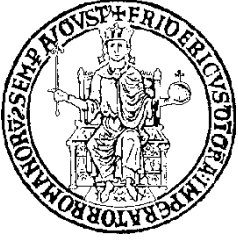
The Quasispecies equation

Assume **nonoverlapping generations**

Mutation matrix : $Q := (Q_{gg'})$: probability that a genotype g is generated in the attempt to reproduce a genotype g'

$$0 \leq Q_{gg'} \leq 1 \quad \sum_g Q_{gg'} = 1, \quad \forall g'$$

Fitness : $W_g \geq 0$ *proportional* to expected number of offspring of an individual of genotype g

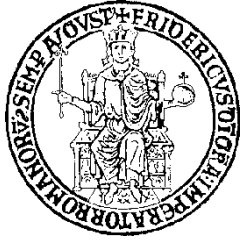


Evolution equation

Genotype fractions $x_g(t) = n_g(t) / \sum_{g'} n_{g'}(t)$

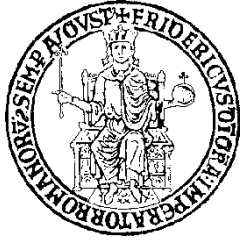
$$x_g(t+1) = \frac{1}{\langle W \rangle_t} \sum_{g'} Q_{gg'} W_{g'} x_{g'}(t)$$

$$\langle W \rangle_t = \sum_g W_g x_g(t)$$



Asymptotic behavior

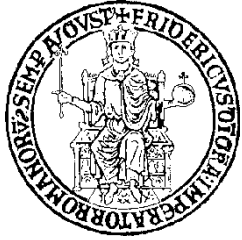




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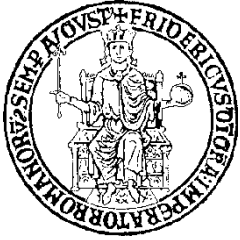


$$x_{\infty}^* = \lim_{t \rightarrow \infty} x_g(t)$$



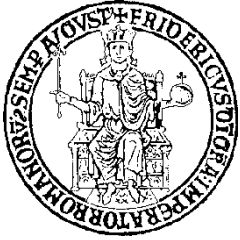
Asymptotic behavior

$$\begin{aligned}x_g^* &= \lim_{t \rightarrow \infty} x_g(t) \\y_g(0) &:= x_g(0), \quad \forall g \\y_g(t+1) &:= \sum_{g'} Q_{gg'} W_{g'} y_{g'}(t), \quad t \geq 0\end{aligned}$$



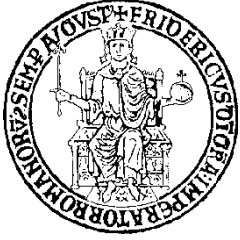
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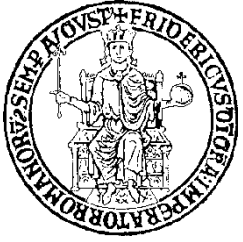
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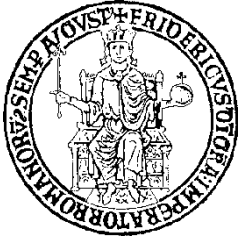
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Thus





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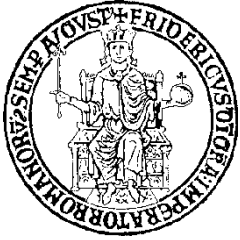
$$\vec{y}(t) \longrightarrow \lambda(t) \vec{y}^*$$



Thus

$$\vec{y}(t) \longrightarrow \lambda(t) \vec{y}^*$$

$$\vec{y}^* : \mathsf{T} \vec{y}^* = \lambda_{\max} \vec{y}^*$$

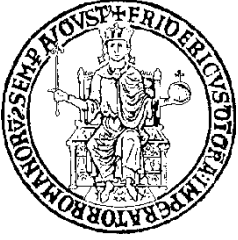


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λ_{\max} : leading eigenvalue of T (positive and non-degenerate)



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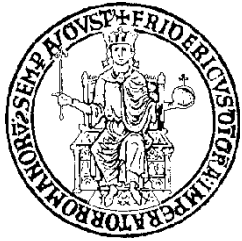
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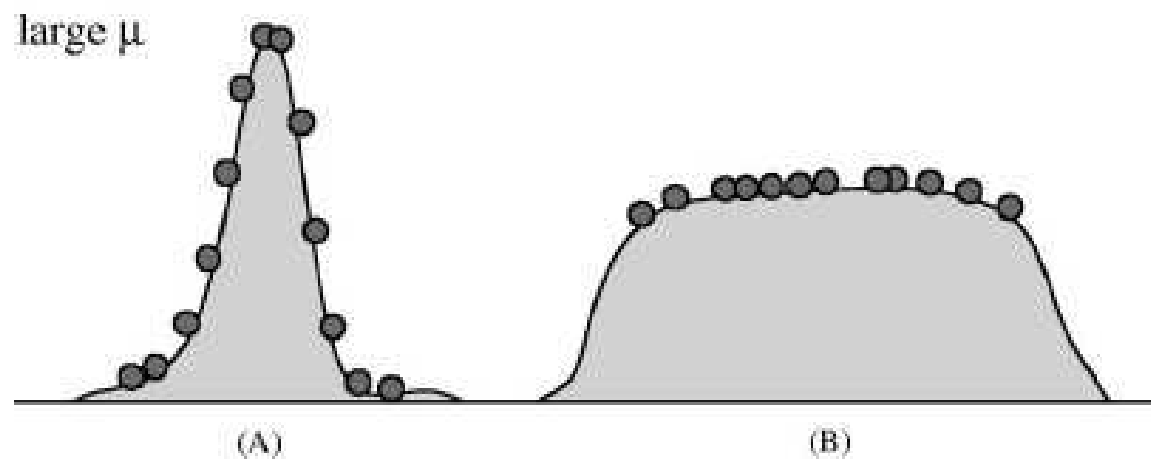
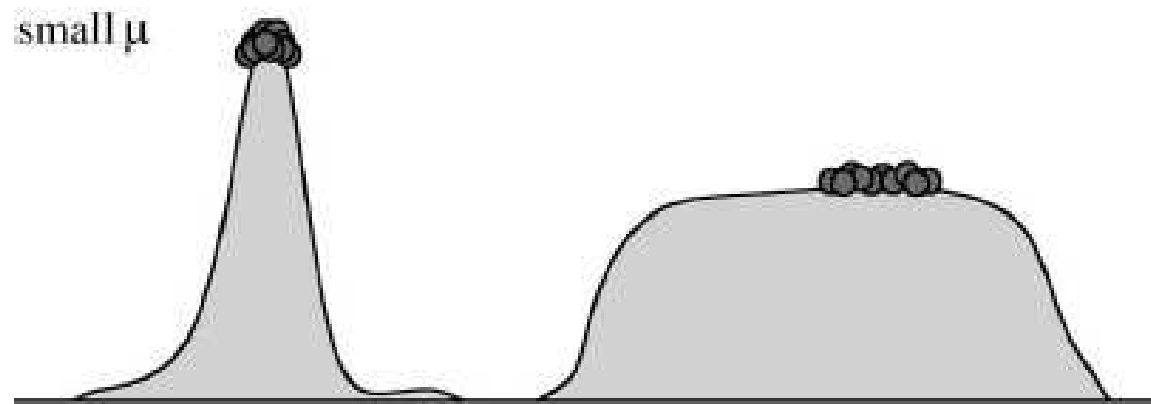
$\log \lambda_{\max}$ looks like a (negated) free energy : fitness + “entropy”

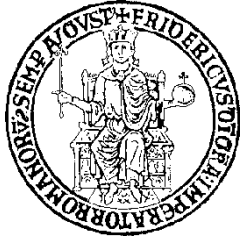


Survival of the flattest

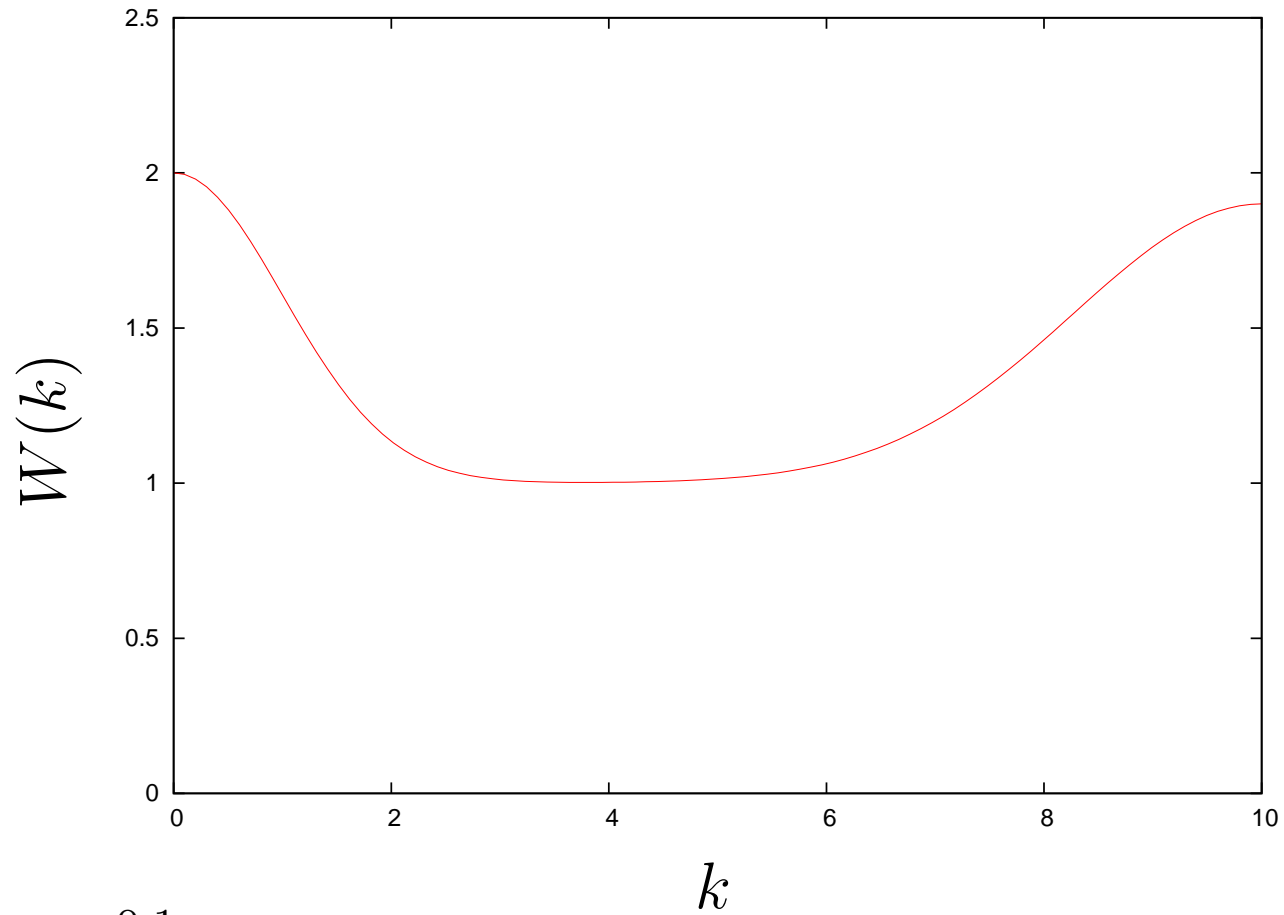


Schuster and Swetina, 1988

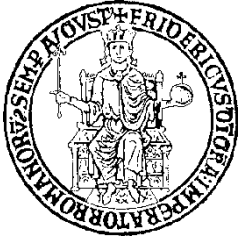




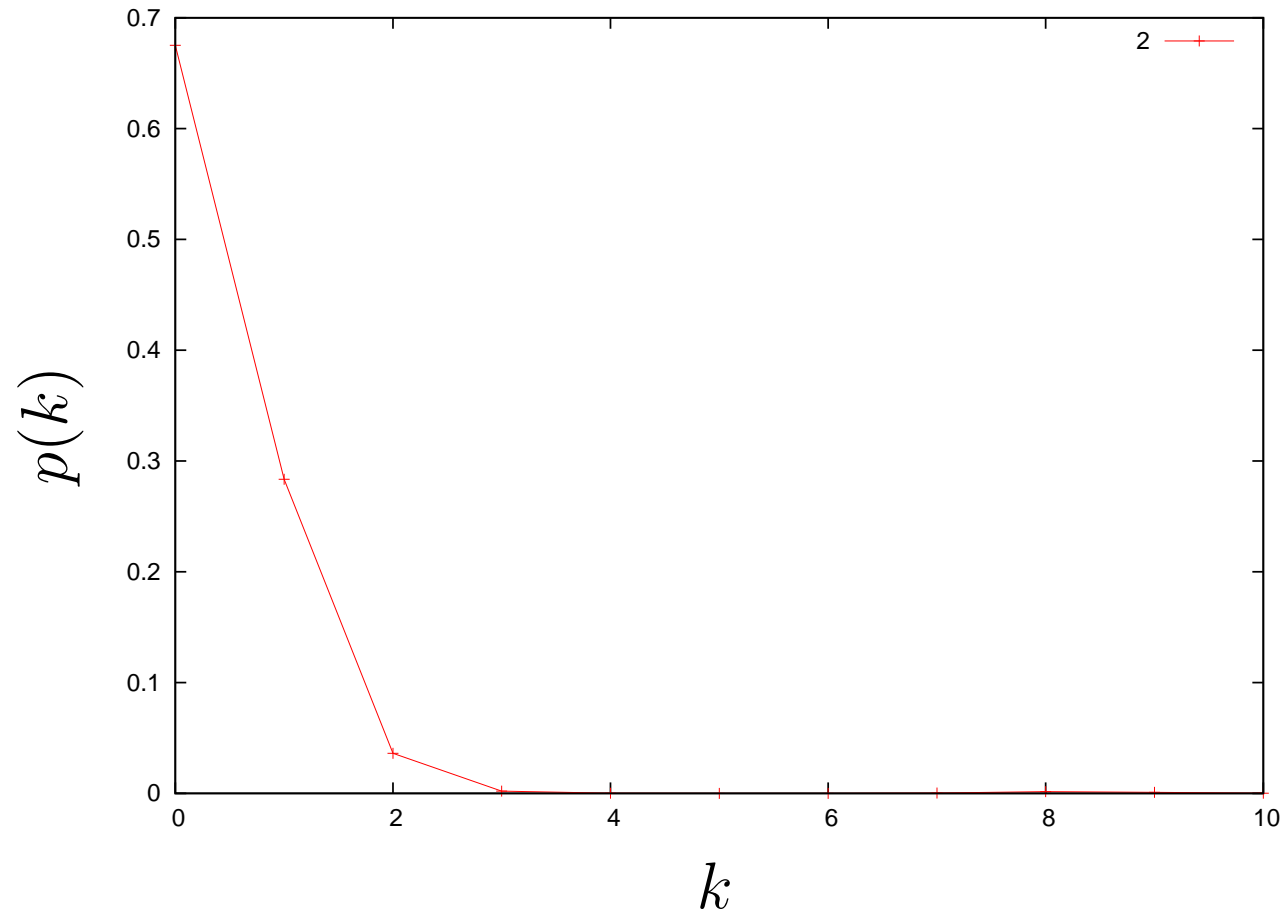
Competition of two peaks

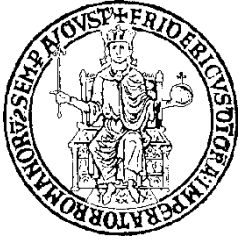


$$\mathcal{A} = 2 \quad \mu = 0.1$$

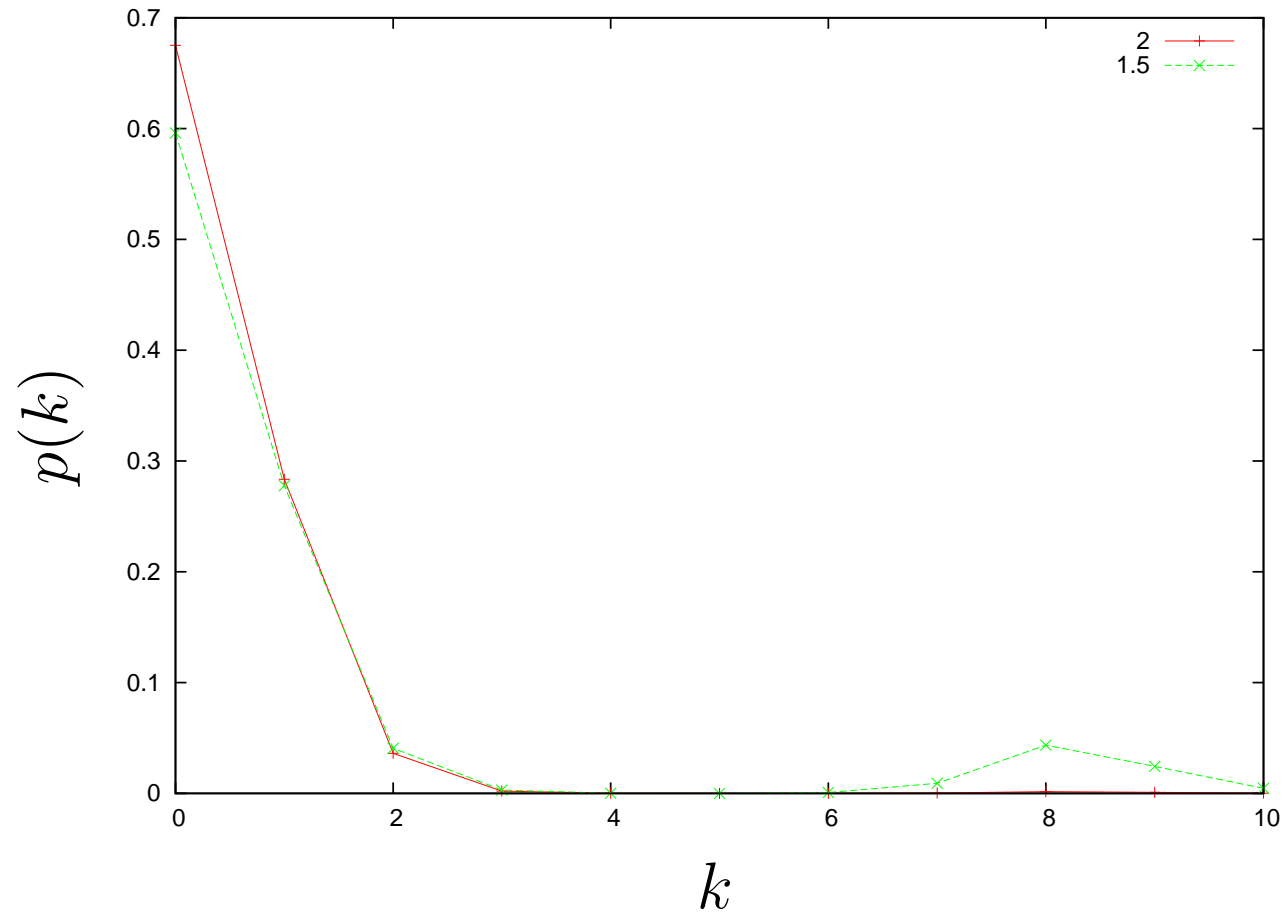


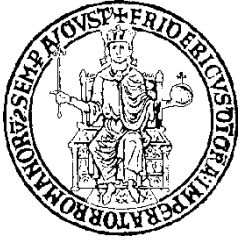
Distribution



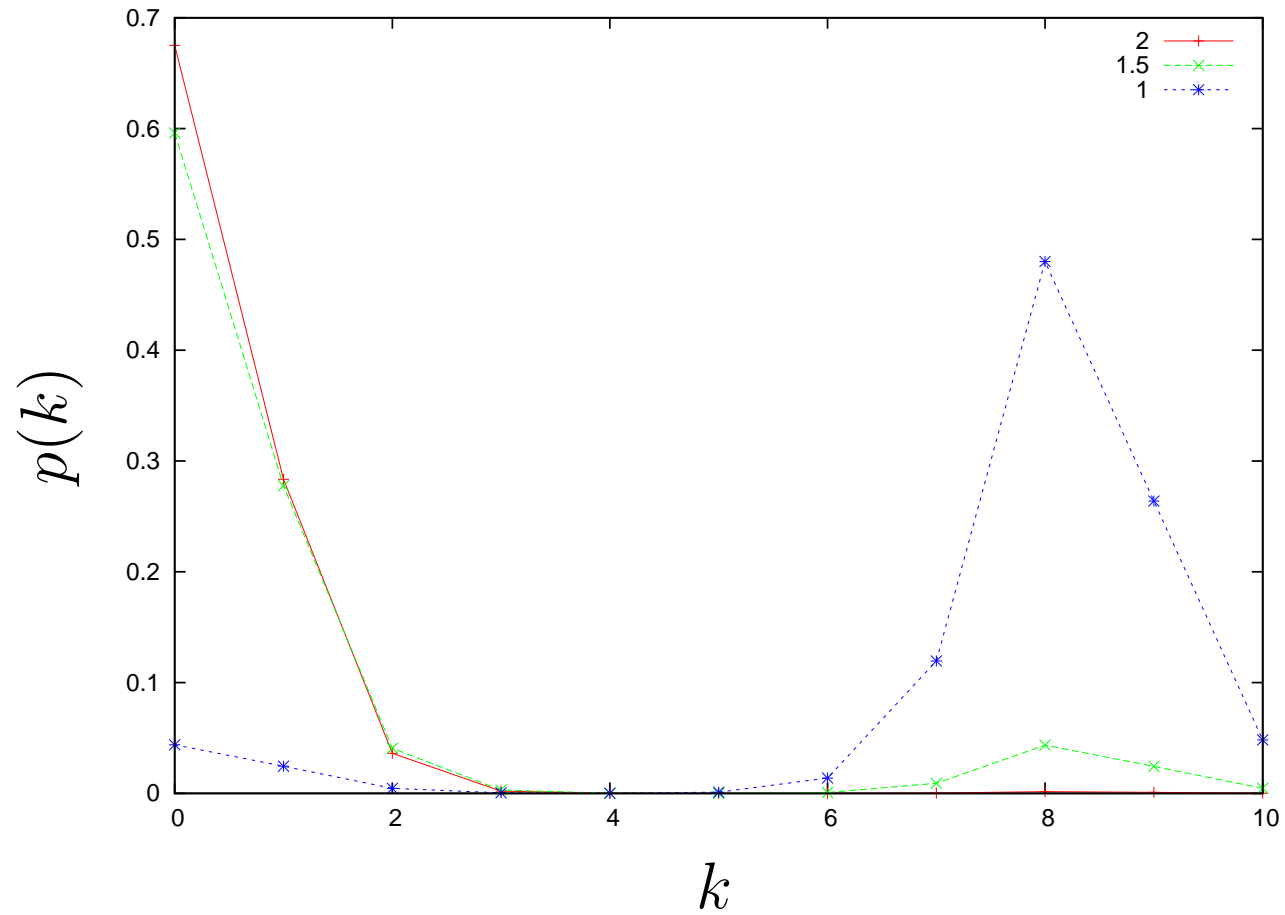


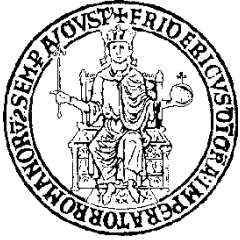
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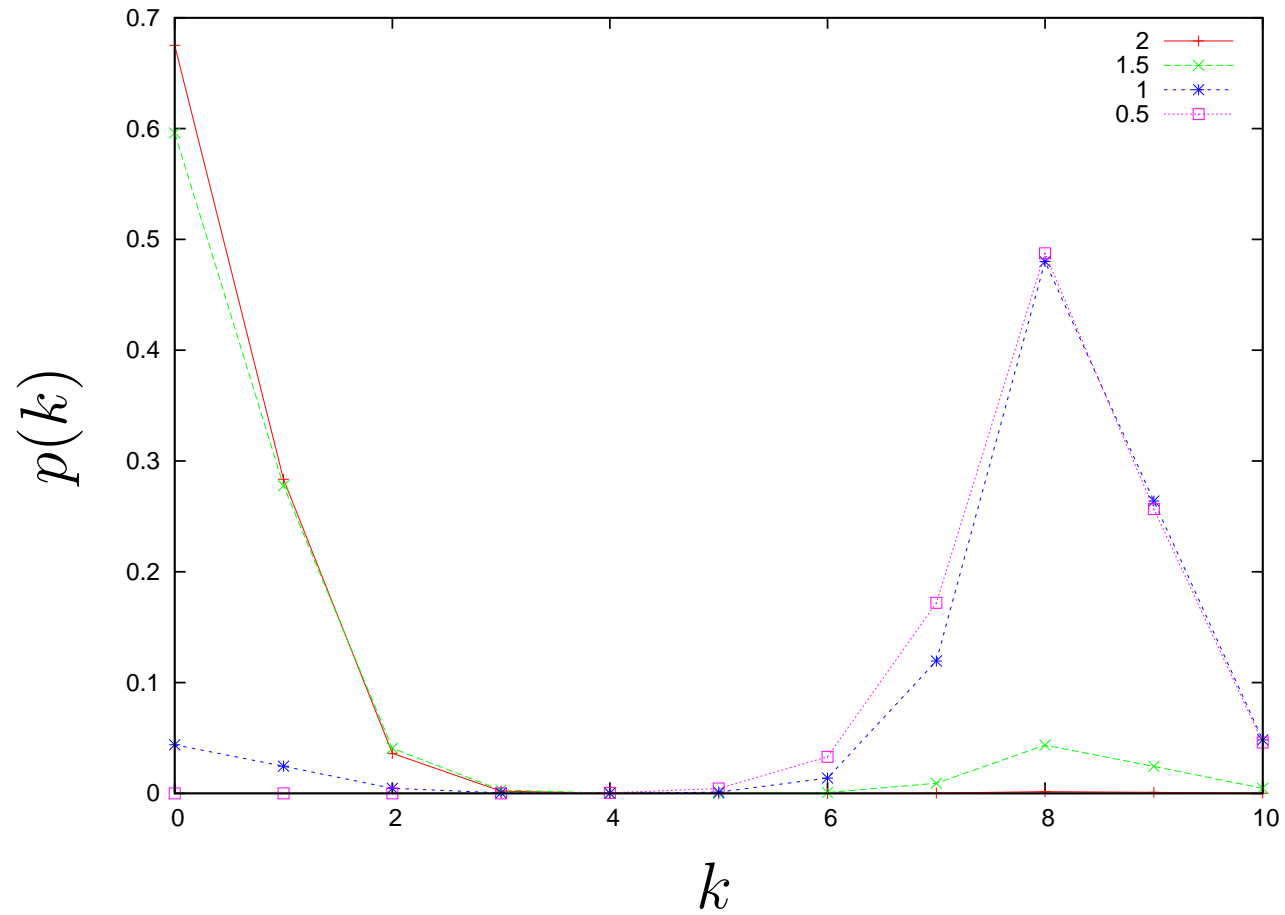


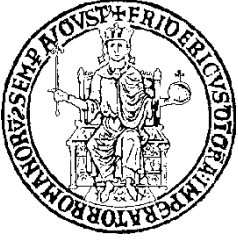
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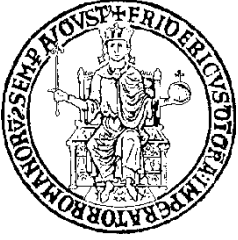


Neutral Networks

Can “entropy” alone drive the evolutionary process ?

E. van Nimwegen, J. F. Crutchfield, M. Huynen, PNAS, 96, 9716 (1999)

Neutral network : $G \subset \Sigma$



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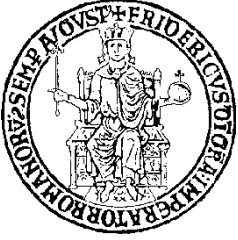
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⑥ Only point mutations are allowed :

$Q_{gg'} = \mu / ((\mathcal{A} - 1)L) \neq 0$, iff g and g' differ in only one site



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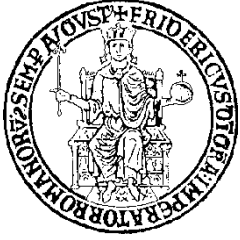
ν_g : probability that the offspring of $g \in G$ belongs to G

d_g : number of single-site mutants of g that belong to G

P : fraction of individuals which lie on G

R : fraction of offspring of individuals *not* belonging to G

that are in G



Evolution in the QS limit

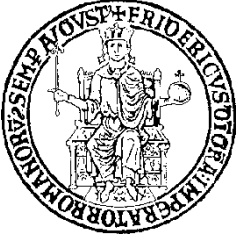
$$P' = \frac{F}{\langle W \rangle} \langle \nu \rangle P + R$$

As $R \ll P$ the steady state satisfies

$$\frac{F}{\langle W \rangle} \langle \nu \rangle = 1$$

Equation relating ν_g and d_g :

$$\nu_g = 1 - \mu \left(1 - \frac{d_g}{(\mathcal{A} - 1)L} \right)$$



Average neutrality

$\vec{P} = (P_g)$: fraction of individuals having genome g in the steady state

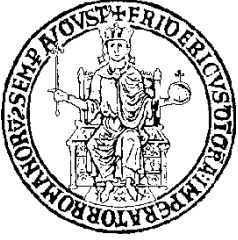
$$P = \sum_{g \in G} P_g$$

Average neutrality :

$$\langle d \rangle = \frac{1}{P} \sum_g d_g P_g$$

From the properties of the steady state we obtain

$$\langle d \rangle = \frac{(\mathcal{A} - 1)L}{\mu} \left(\mu - \frac{F - \langle W \rangle}{F} \right)$$



Neutrality and network topology

$$[g]_G := \{g' \in G, g' \text{ n.n. of } g\}$$

Equation for P_g :

$$P_g = (1 - \mu) \frac{F}{\langle W \rangle} P_g + \frac{\mu}{(\mathcal{A} - 1)L \langle W \rangle} \sum_{g' \in [g]_G} F P_{g'}$$

Adjacency matrix $G = (G_{gg'})$:

$$G_{gg'} = \begin{cases} 1, & \text{if } g \in [g']_G \\ 0, & \text{otherwise} \end{cases}$$

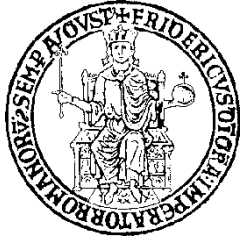


$\langle d \rangle$ **and** G

Using the expression of $\langle d \rangle$ we obtain

$$\langle d \rangle \vec{P} = G \cdot \vec{P}$$

Thus (for a *connected* network) $\langle d \rangle$ is equal to the leading eigenvalue ρ of the adjacency matrix and \vec{P} is the corresponding eigenvector



Finite-size populations

Population of N individuals, uniform fitness
Probability that two individuals do not have the same parent :

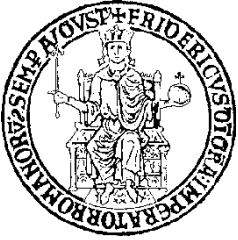
$$\Pi = \left(1 - \frac{1}{N}\right)$$

Probability that two individuals did not have the same ancestor t generations ago :

$$\Pi_t = \Pi \Pi_{t-1} = \Pi^t \simeq e^{-t/N}$$

Probability that the last common ancestor of two individuals lived t generations ago :

$$p(t) = -\frac{d\Pi_t}{dt} = \frac{1}{N} e^{-t/N}$$



Neutral evolution

Average number of mutations between two individuals

$$\langle m \rangle = \int_0^{\infty} dt \, 2\mu t p(t) = 2\mu N$$

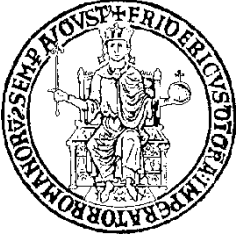
Average number of mutations between any present individual and an individual t generations ago :

$$D(t) = \int_0^{\infty} dt' \, \mu(2t' + t) \frac{1}{N} e^{-t'/N} = \mu(2N + t)$$

Effective mutation rate (Kimura's theorem) :

$$\mu_{\text{eff}} := \frac{dD}{dt} = \mu$$

Finite population on a neutral network

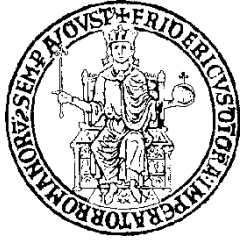


“Blind” ant in a labyrinth : Chooses a neighbor at random, if it's fit moves to it

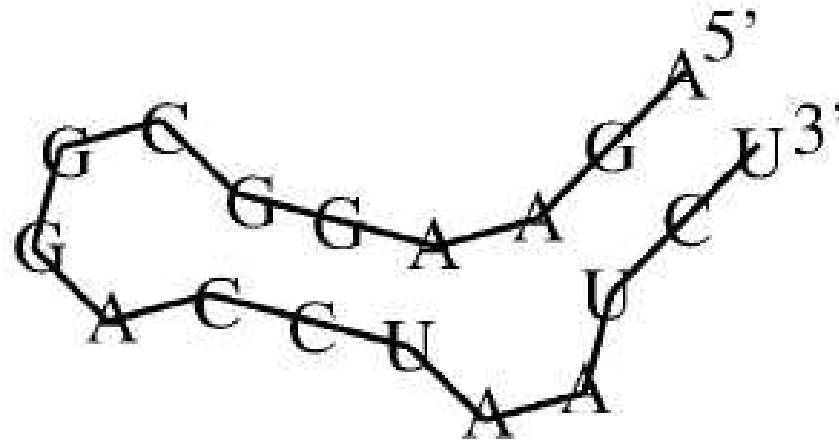
$$P_g^{\text{eq}} = \text{const.} \quad \Rightarrow \quad \langle d \rangle = \frac{1}{|G|} \sum_{g \in G} d_g = \bar{d}$$

“Myopic” ant in the labyrinth : Chooses at random one among its fit neighbors

$$P_g^{\text{eq}} \propto d_g \quad \Rightarrow \quad \langle d \rangle = \frac{1}{|G|\bar{d}} \sum_{g \in G} d_g^2 = \hat{d}$$



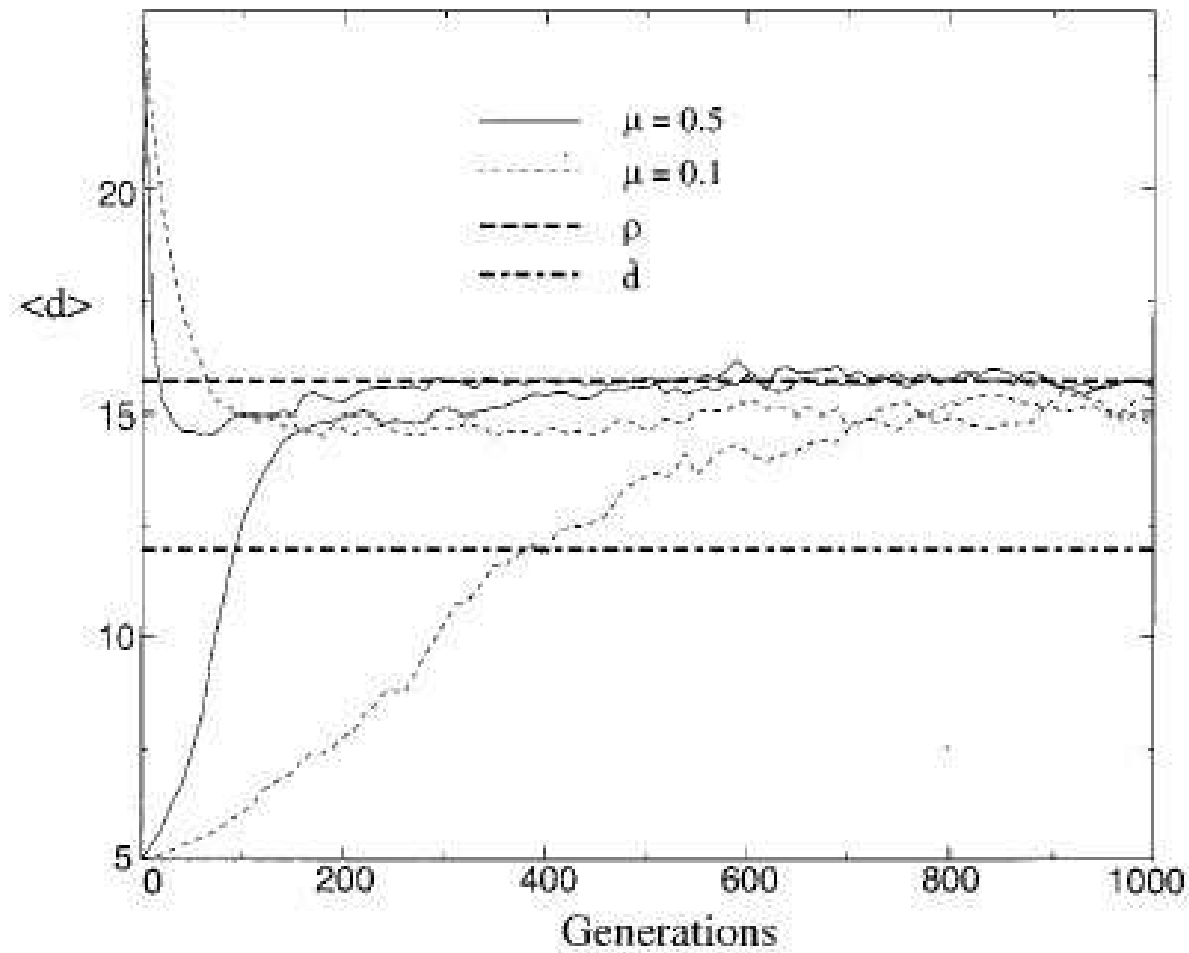
RNA Secondary structure

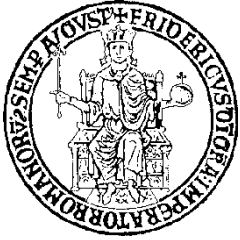


Neutral network composed by 51,028 sequences

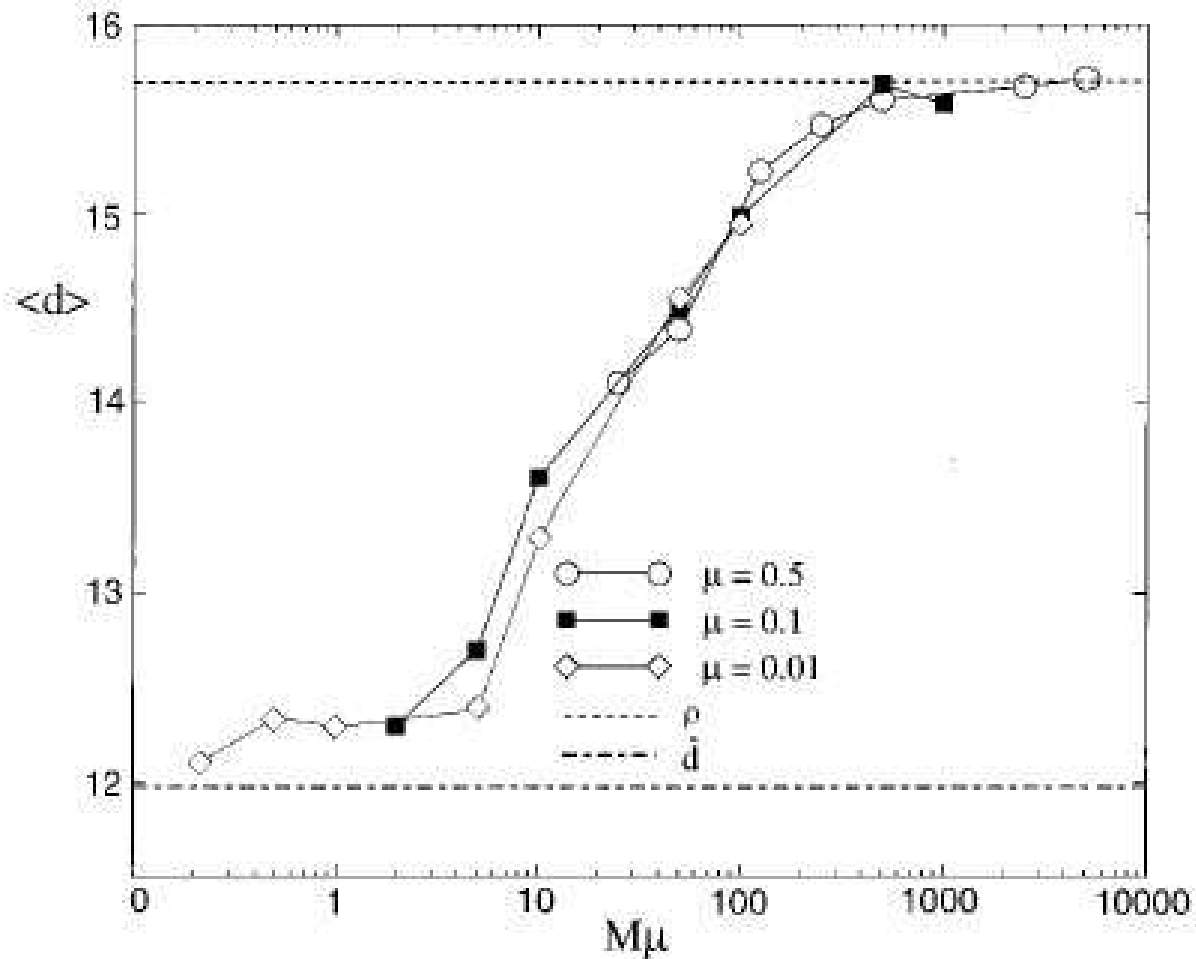


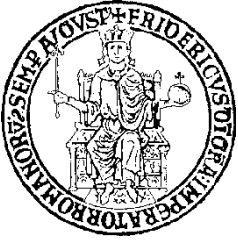
Evolution of robustness





Robustness at equilibrium





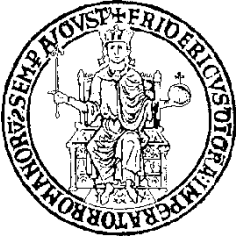
How neutral is neutral ?

Population of size N , two genotypes a and b

$$\frac{W_a}{W_b} = e^s$$

$\mathcal{P}(x, t)$: pdf that $x_a = x$ at time t Kimura's diffusion approximation (t continuous, $N \gg 1$)

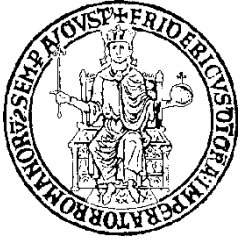
$$\frac{\partial \mathcal{P}}{\partial t} = \frac{1}{2N} \frac{\partial^2}{\partial x^2} \{x(1-x) \mathcal{P}\} - s \frac{\partial}{\partial x} \{x(1-x) \mathcal{P}\}$$



Equations at the boundary ($s \ll 1$) :

$$\frac{\partial \mathcal{P}(0, t)}{\partial t} \simeq \frac{1}{N} \lim_{x \rightarrow 0^+} \mathcal{P}(x, t)$$
$$\frac{d\mathcal{P}(1, t)}{dt} \simeq \frac{1}{N} \lim_{x \rightarrow 1^-} \mathcal{P}(x, t)$$

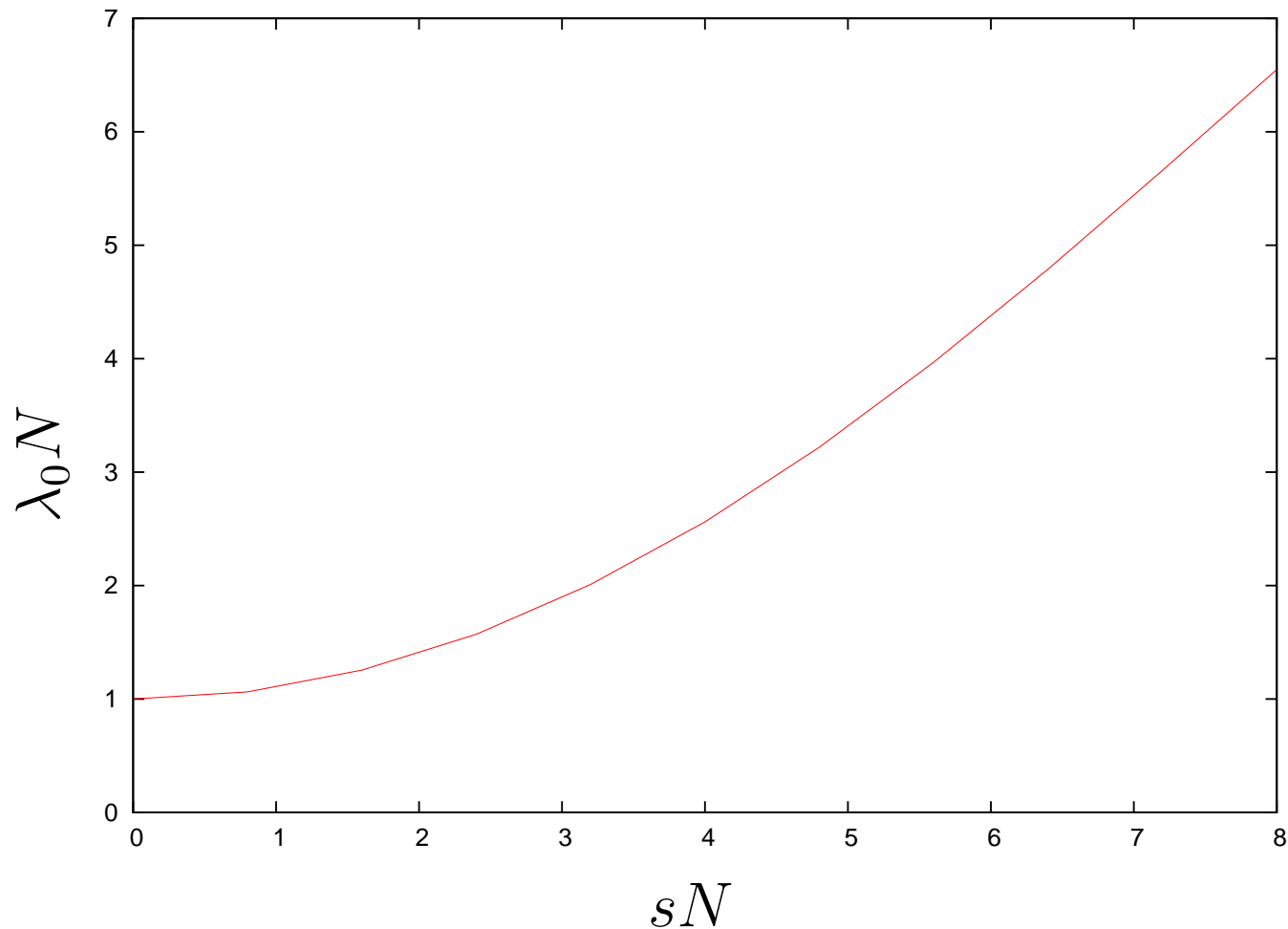
The probability that $x \notin \{0, 1\}$ decays like $e^{-\lambda_0 t}$, where λ_0 is the minimal eigenvalue

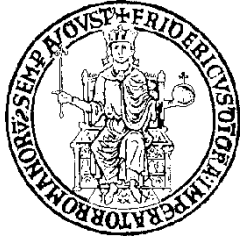


Selection rate

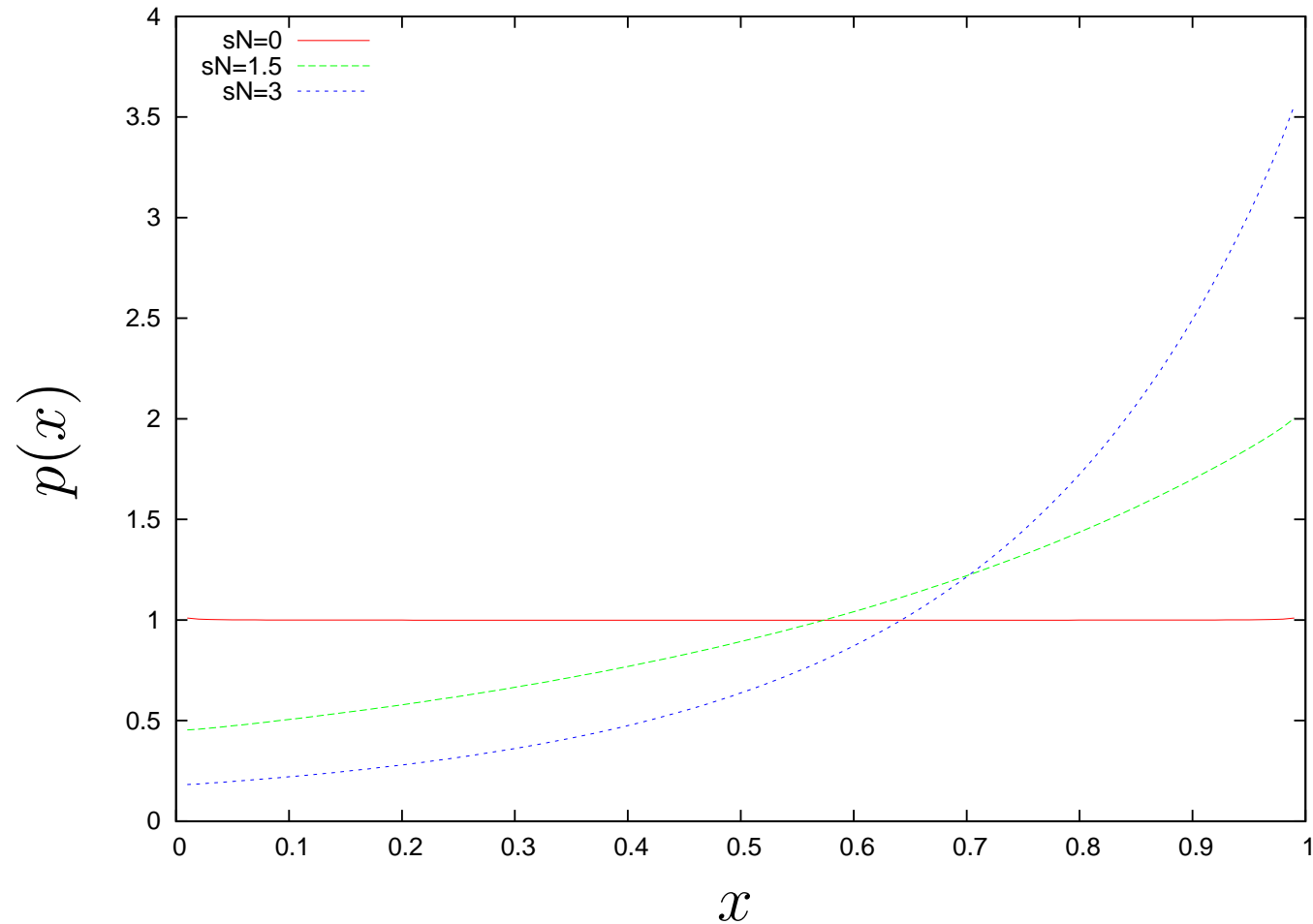


$N\lambda_0$ is a function of sN :

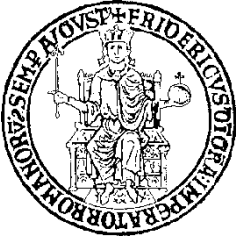




Frequency distribution



Frequency distribution in *unfixed* classes vs. sN



The backward diffusion equation

Let x be given and the initial condition x_0 independent.
Then

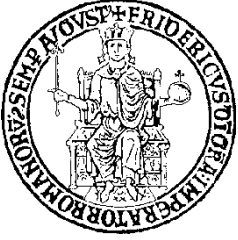
$$\frac{\partial}{\partial t} \mathcal{P}(x, t | x_0, t_0) = \frac{x_0(1-x_0)}{2N} \frac{\partial^2 \mathcal{P}}{\partial x_0^2} + s x_0(1-x_0) \frac{\partial \mathcal{P}}{\partial x_0}$$

Fixation probability by time t : $\phi(x_0, t) = \mathcal{P}(1, t | x_0, 0)$

Ultimate fixation probability : $\phi(x_0) = \lim_{t \rightarrow \infty} \phi(x_0, t)$

$$\frac{1}{2N} \frac{d^2 \phi}{dx_0^2} + s \frac{d\phi}{dx_0} = 0$$

Boundary conditions : $\phi(0) = 0$; $\phi(1) = 1$



Kimura-Ohta fixation probability

$$\phi(x_0) = \frac{1 - e^{-2N_e s x_0}}{1 - e^{-2N_e s}}$$

Rate of fixation of a mutant with selective advantage s
($x_0 = 1/N$) :

$$u = \mu N_e \frac{1 - e^{-2s}}{1 - e^{-2N_e s}}$$

$$u \simeq \begin{cases} 0, & \text{for } N_e s \lesssim -1 \text{ (deleterious mutations)} \\ \mu, & \text{for } N_e |s| \ll 1 \text{ (neutral mutations)} \\ 2\mu N_e s, & \text{for } N_e s \gtrsim 1 \text{ (beneficial mutations)} \end{cases}$$



Note : *Effective population size*

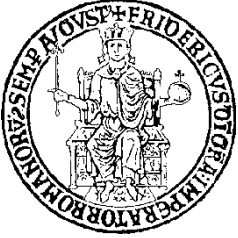
N_e : *effective* population size

$$\frac{1}{N_e} = \frac{1}{t} \int_0^t dt' \frac{1}{N(t')}$$

With exponential growth

$$N_e(t) \simeq N_0 \log \frac{N(t)}{N_0}$$

For humans ($N_0 \simeq 10\,000$, $N(t) \simeq 6 \cdot 10^9$) $N_e \simeq 133\,000$



Balance relation

J. Berg, S. Willmann, M. Lässig, BMC Evol. Biol., 4, 42 (2004)

$u_{gg'}$: probability that mutant g reaches fixation in a population of g'

$$\frac{u_{gg'}}{u_{g'g}} = e^{2(N-1)s} \simeq e^{2Ns}$$

where $s = \Delta \ln W = \Delta F$



Evolving monomorphic population

$P_g(t)$: Probability of finding genotype g fixated in an *ensemble* of populations

$Q_{gg'}$: Probability that a mutation of genotype g' produces a genotype g

For a *reversible* mutation process one has (P_g^0 is the *a priori* probability of g)

$$\frac{Q_{gg'}}{Q_{g'g}} = \frac{P_{g'}^0}{P_g^0}$$



Equilibrium distribution

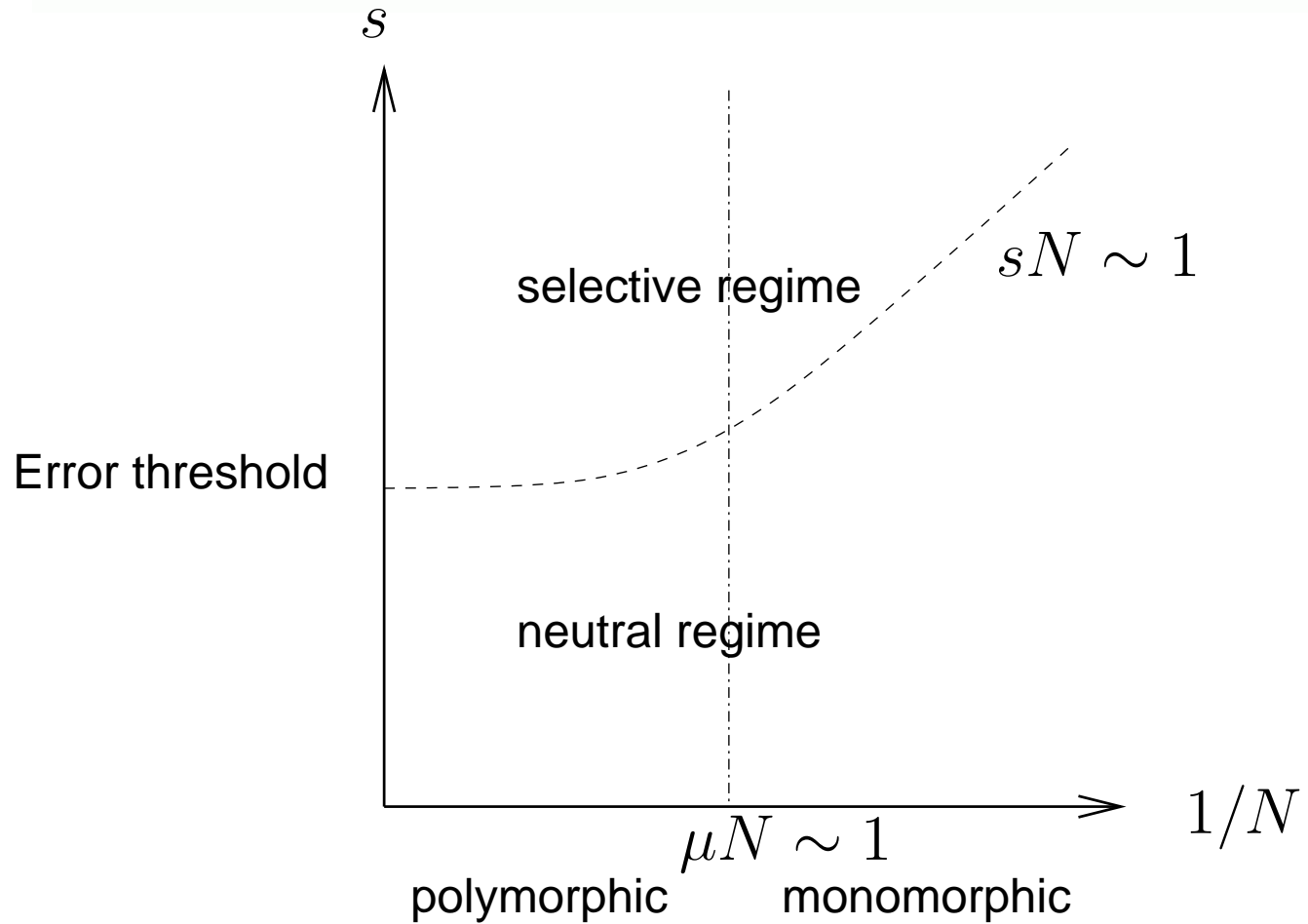
$$\frac{\partial P_g}{\partial t} = \sum_{g' (\neq g)} [u_{gg'} Q_{gg'} P_{g'} - u_{g'g} Q_{g'g} P_g]$$

From detailed balance :

$$P_g^{\text{eq}} = P_g^0 e^{2NF}$$

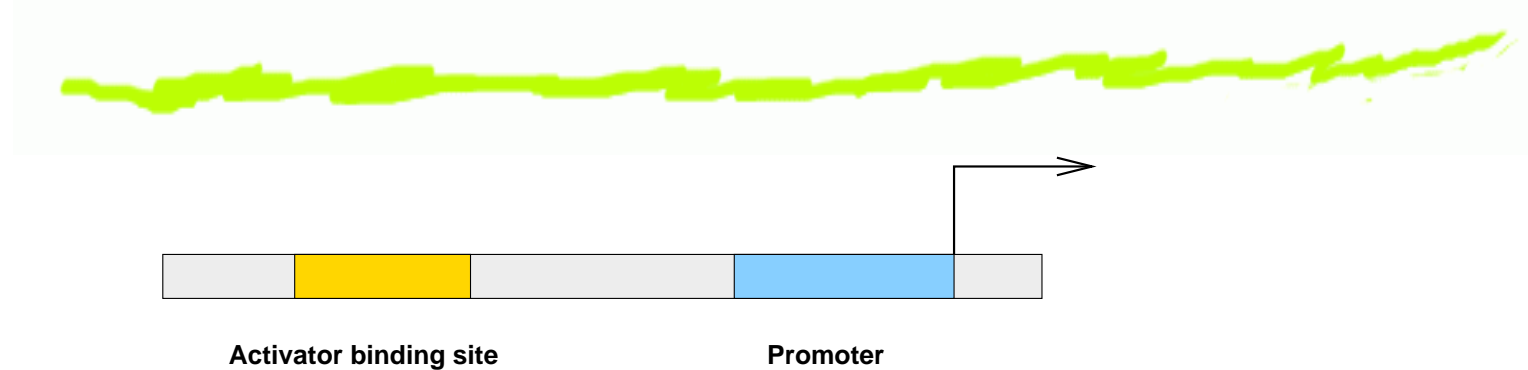
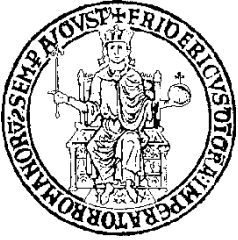


“Phase diagram”



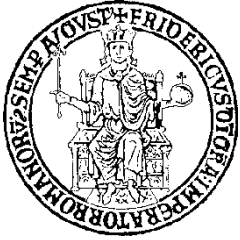
Lässig

Evolution of transcription factor binding sites

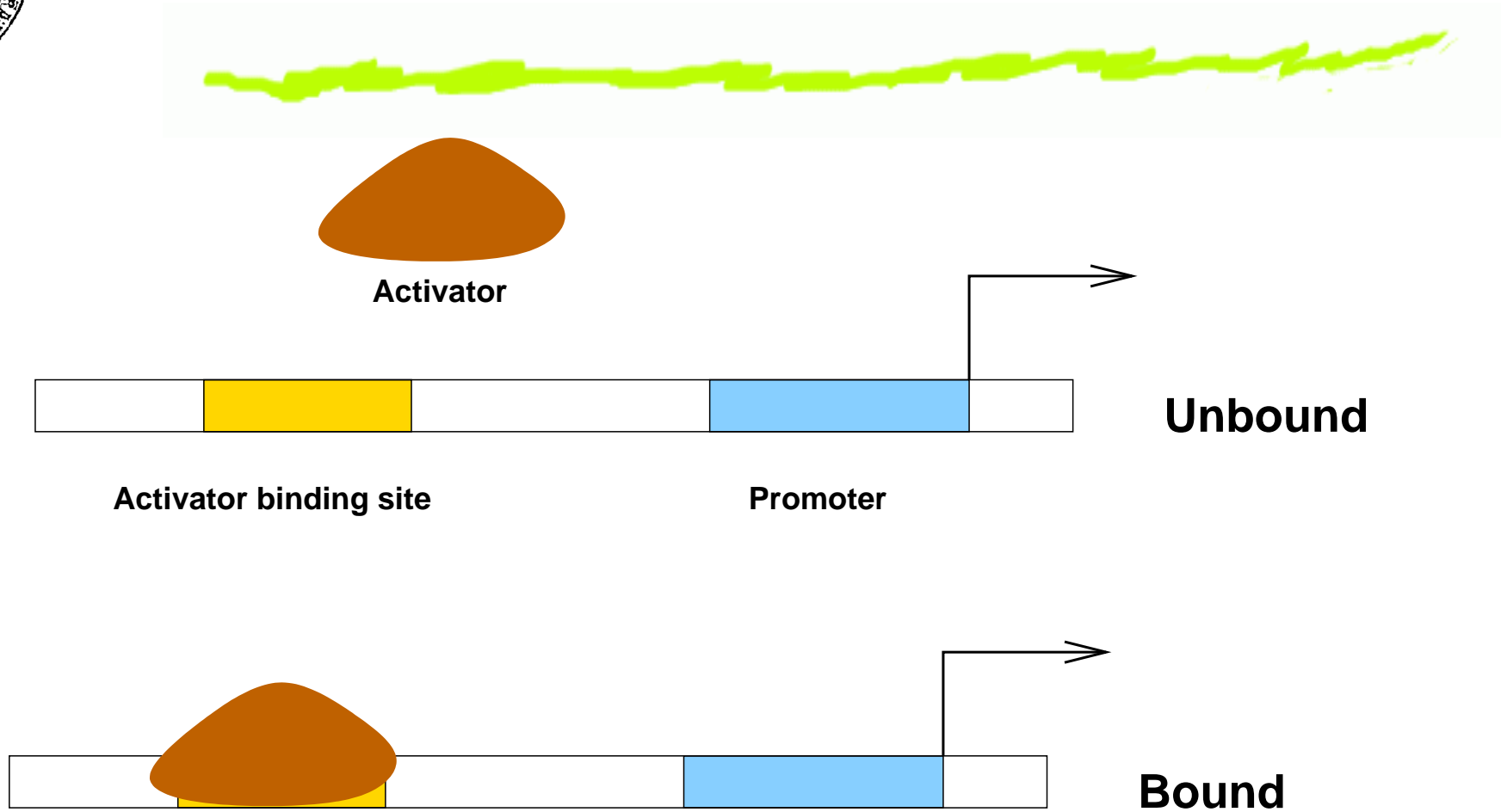


- U. Gerland and T. Hwa, *J. Mol. Evol.*, **55**, 386 (2002) ;
U. Gerland, J. D. Moroz and T. Hwa, *PNAS*, **99**, 12015 (2002) : large populations : quasispecies theory
- A. M. Sengupta, M. Djordjevic and B. Shraiman, *PNAS*, **99**, 2072 (2002) : multiple binding sites
- A. Wagner and P. F. Stadler, *J. Exp. Zool.*, **285**, 119 (1999) : Robustness of viral secondary RNA structures
- J. Berg, S. Willmann and M. Lässig, *BMC Evol. Biol.*, **4**, 42 (2004) : Small populations : Kimura-Ohta random

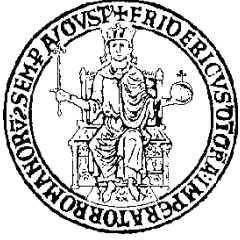
walk



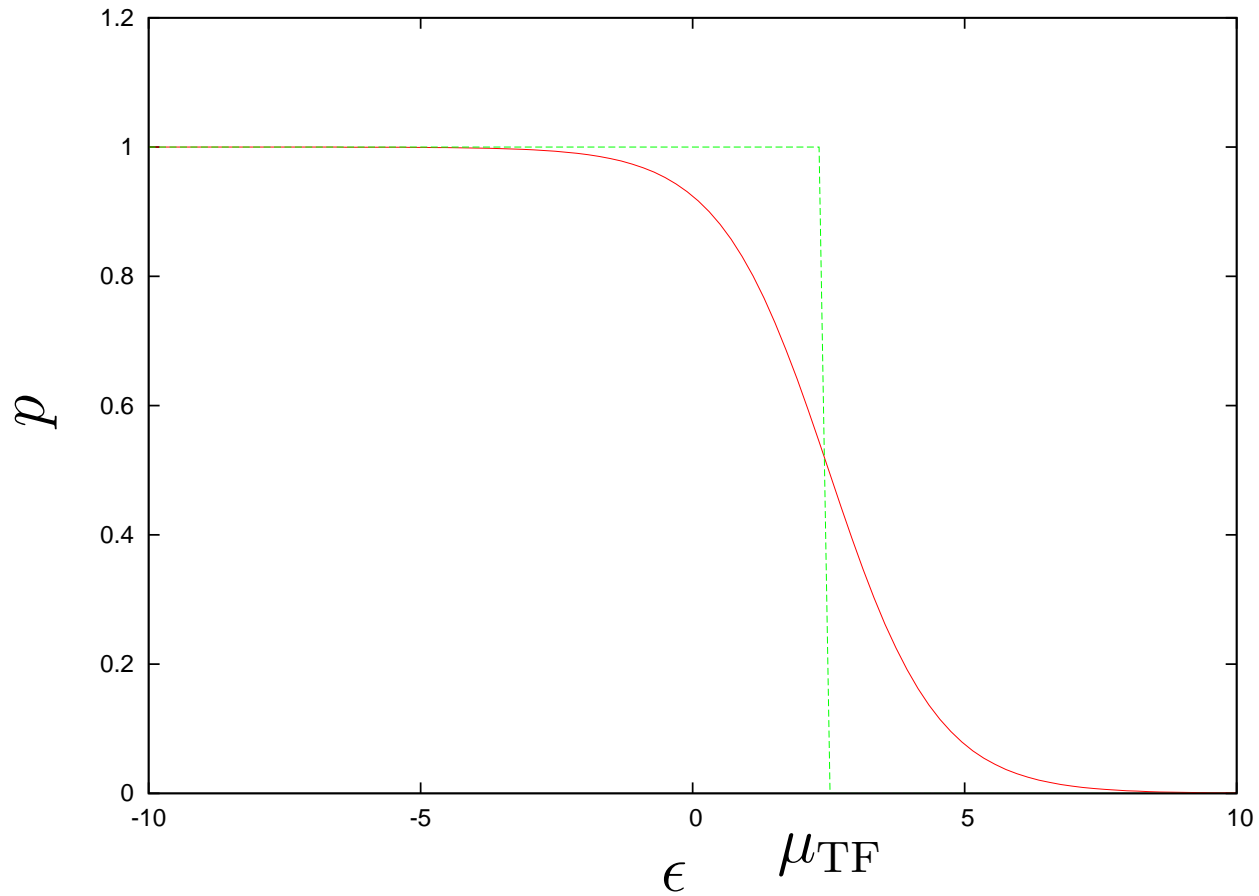
Transcription factor binding



$$p(\text{bound}) = \frac{1}{1 + e^{-(\epsilon - \mu_{\text{TF}})/k_B T}}$$



Binding probability



Binding site affinity : The linear model



G. Stormo and D. Fields, Trends Biochem. Sci., 23, 109 (1998)

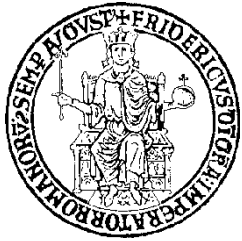
$$\epsilon = \sum_{i=1}^L \epsilon_i(\mathbf{g}_i), \quad \mathbf{g}_i \in \{A, C, G, T\}$$

Two-state model

$$\epsilon \approx \epsilon_0 r = \epsilon_0 \sum_{i=1}^L \left(1 - \delta_{\mathbf{g}_i \mathbf{g}_i^0}\right)$$

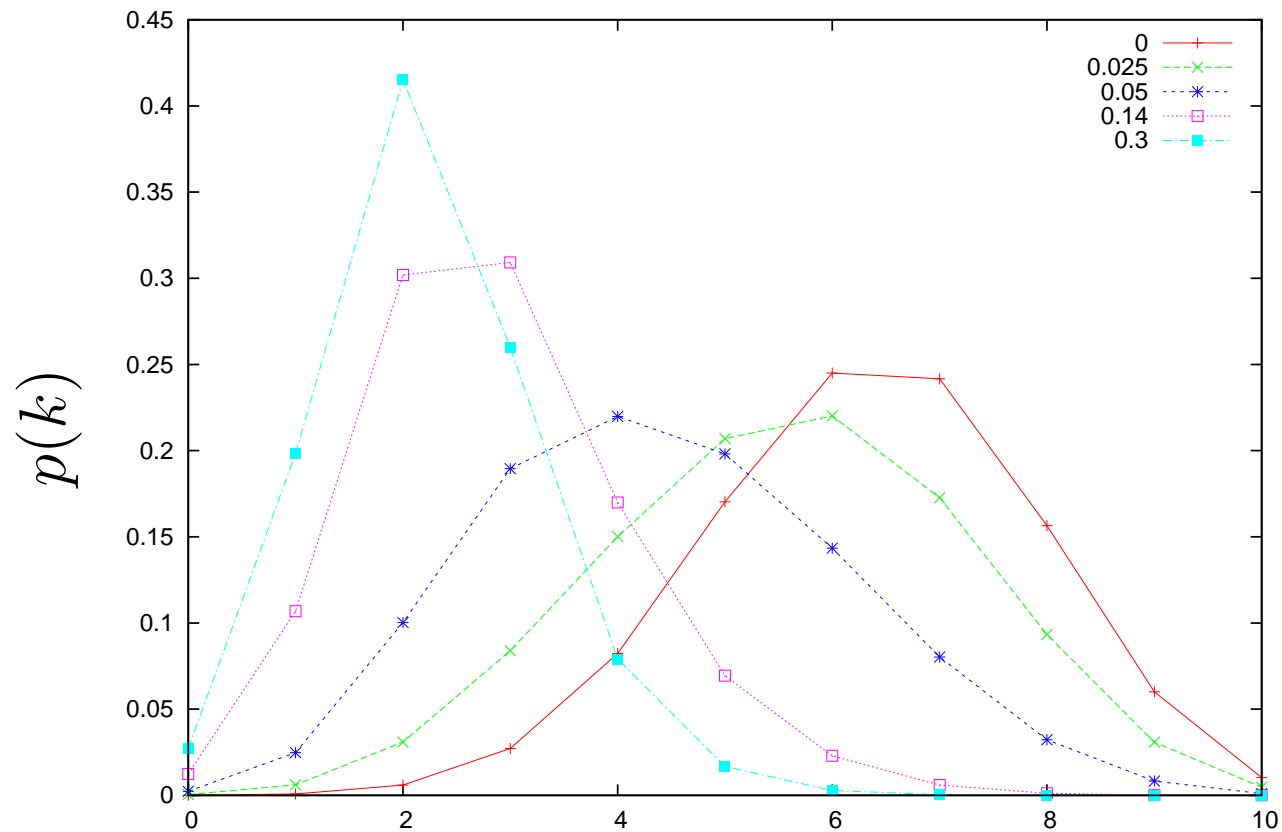
Fitness

$$W(k) \propto 1 + \alpha p(\text{bound})$$

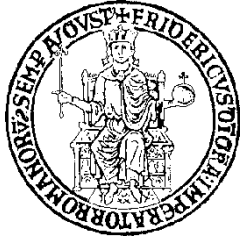


The QS approximation

Very large populations (bacteria)



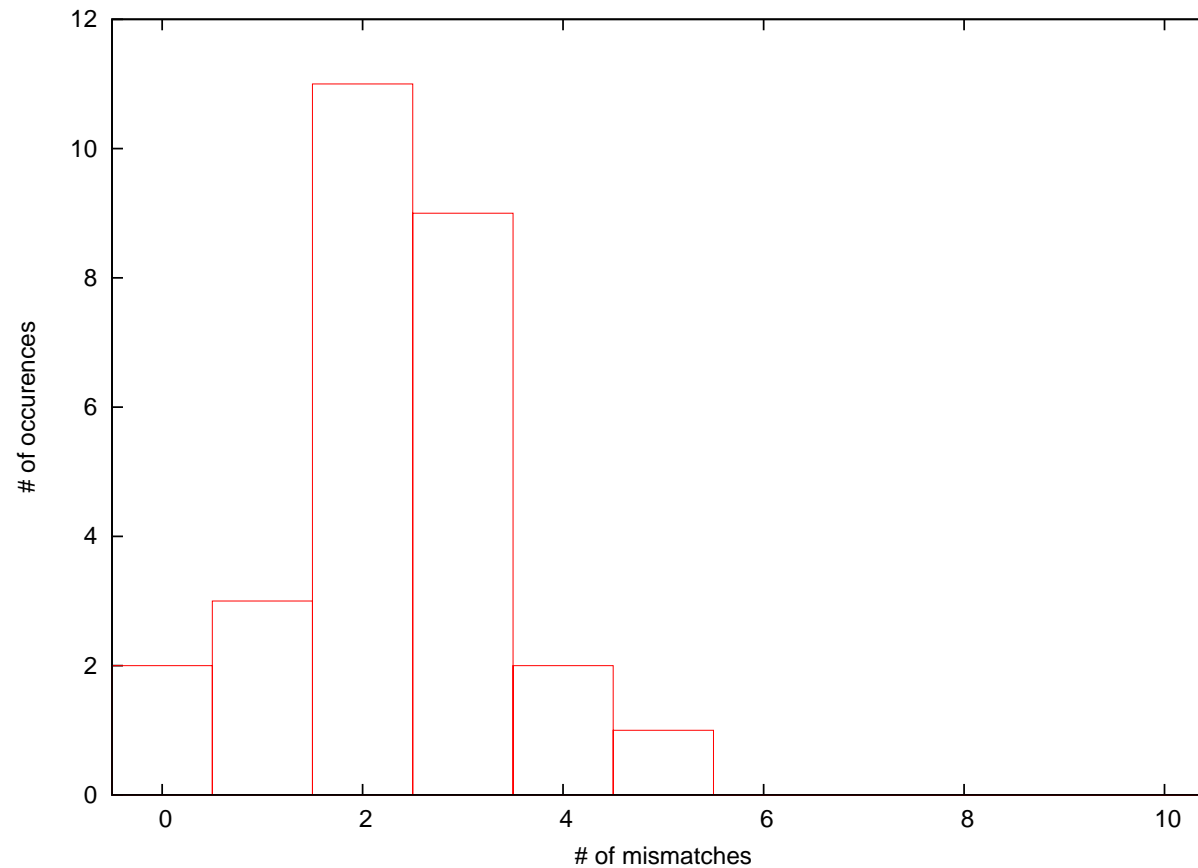
$$\mathcal{A} = 4 \quad \mu = 0.1 \quad \epsilon_0/k_B T = 2 \quad \mu_{\text{TF}} = 3\epsilon_0$$



Empirical distribution

CRP (c-AMP receptor protein) binding sites in *E. Coli*

MH Saier et al., 1996



Robustness optimization for a network



Death rate of binding sites :

$$\gamma = \text{Loss rate} + \text{Rate of birth of spurious sites}$$

Loss rate : $\kappa(\mu_{\text{TF}}) \propto \mu_{\text{TF}}/k_{\text{B}}T$

Rate of birth for spurious states :

$$\kappa_{\text{sp}}(\mu_{\text{TF}}) \propto (\mu_{\text{TF}}/k_{\text{B}}T) e^{-\mu_{\text{TF}}/k_{\text{B}}T}$$

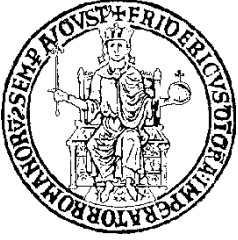
n_i : # of binding sites

N_{cR} : # potential spurious binding sites

Total death rate for N_{F} factors :

$$\gamma = \sum_{i=1}^{N_{\text{F}}} \left[n_i \kappa(\mu_{\text{TF}}^i) + N_{\text{cR}} \kappa_{\text{sp}}(\mu_{\text{TF}}^i) \right]$$

Robustness optimization for a network II



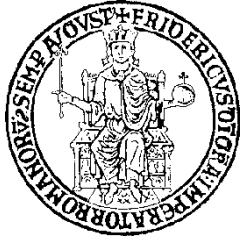
Optimizing γ :

$$n_i / N_{cR} \propto (\mu_{TF}^i / k_B T) e^{-\mu_{TF}^i / k_B T}$$

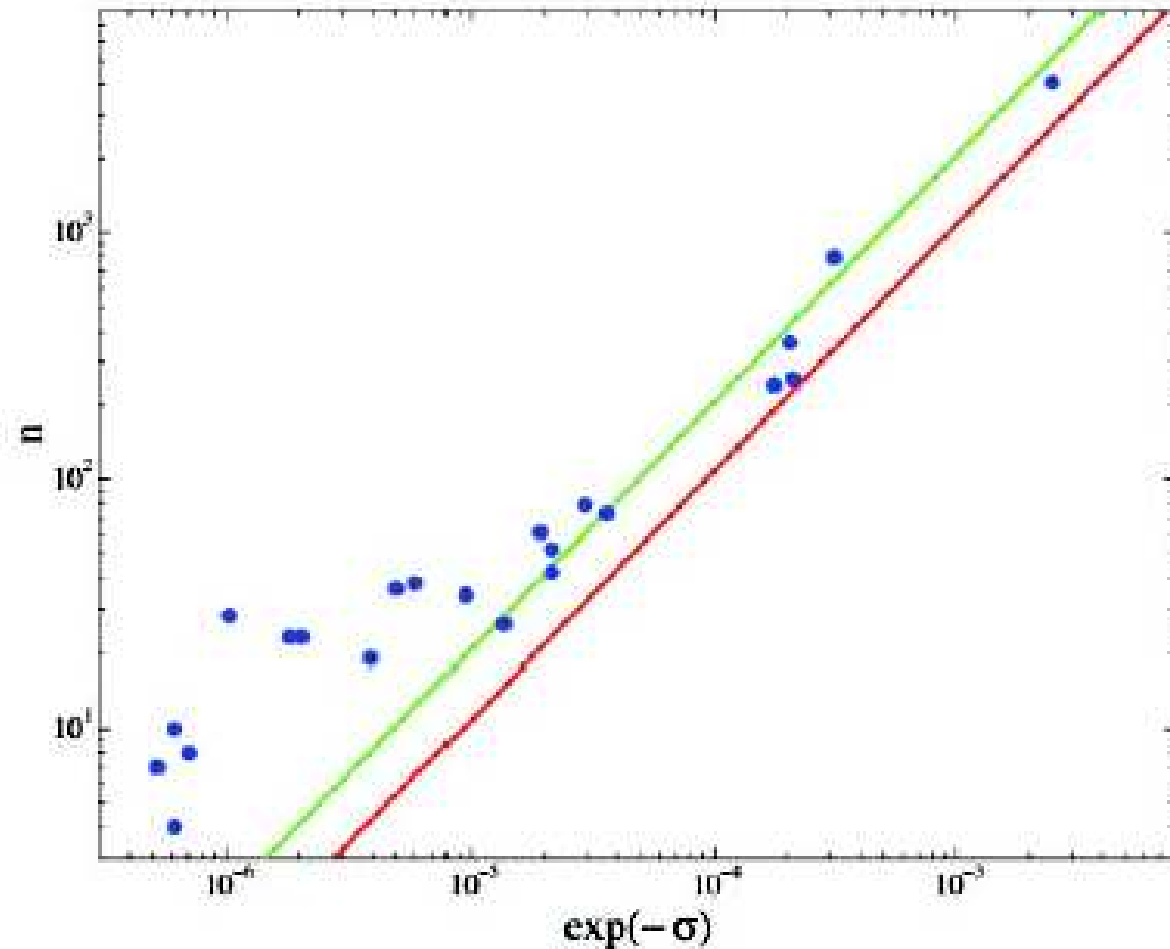
Since

$$e^{-\mu_{TF} / k_B T} \propto [TF] = \rho_i$$

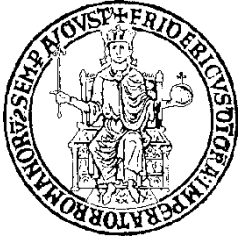
$$\rho_i \propto n_i + \text{logarithmic corrections}$$



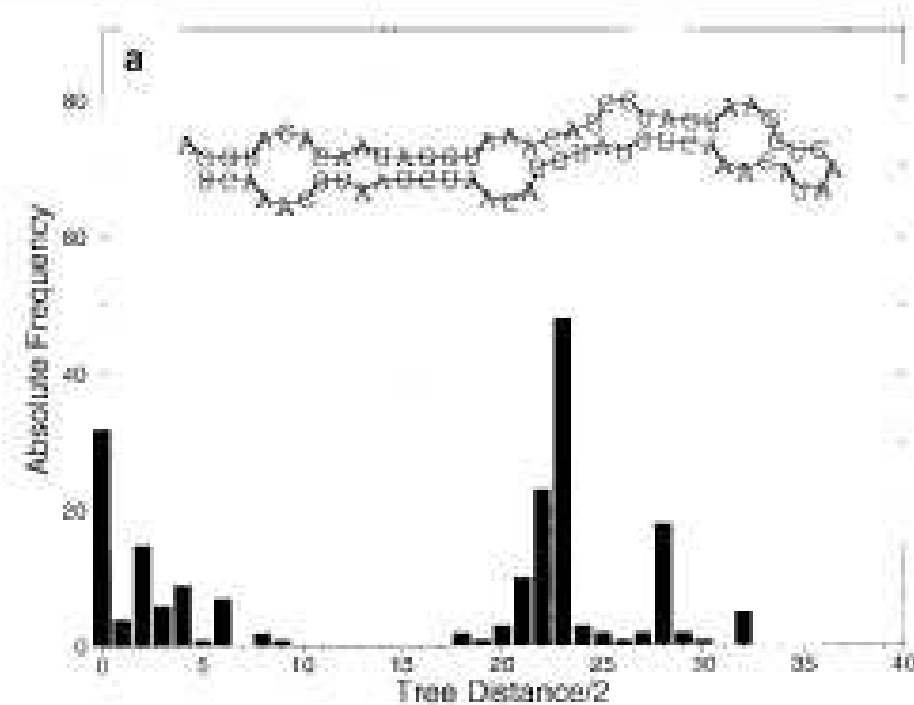
Pleiotropy vs. affinity



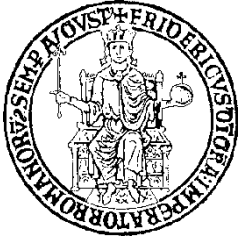
N.B. : $\sigma = \mu_{\text{TF}}/k_{\text{B}}T$



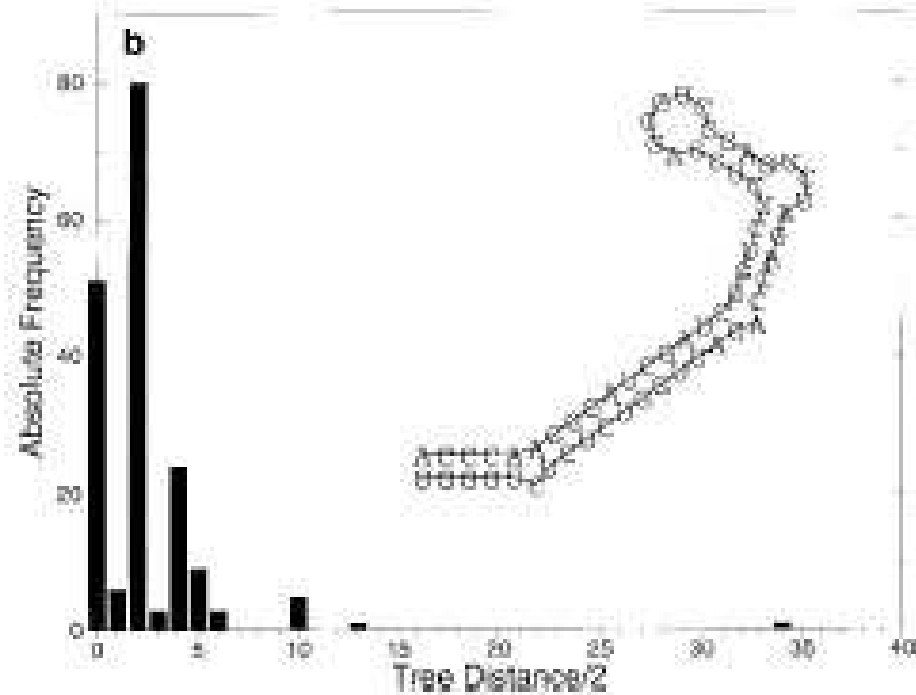
Viral RNA structures : Nonconserved



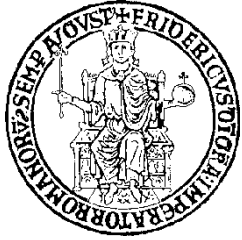
HIV sequence HIVANT70, pos. 7089–7155



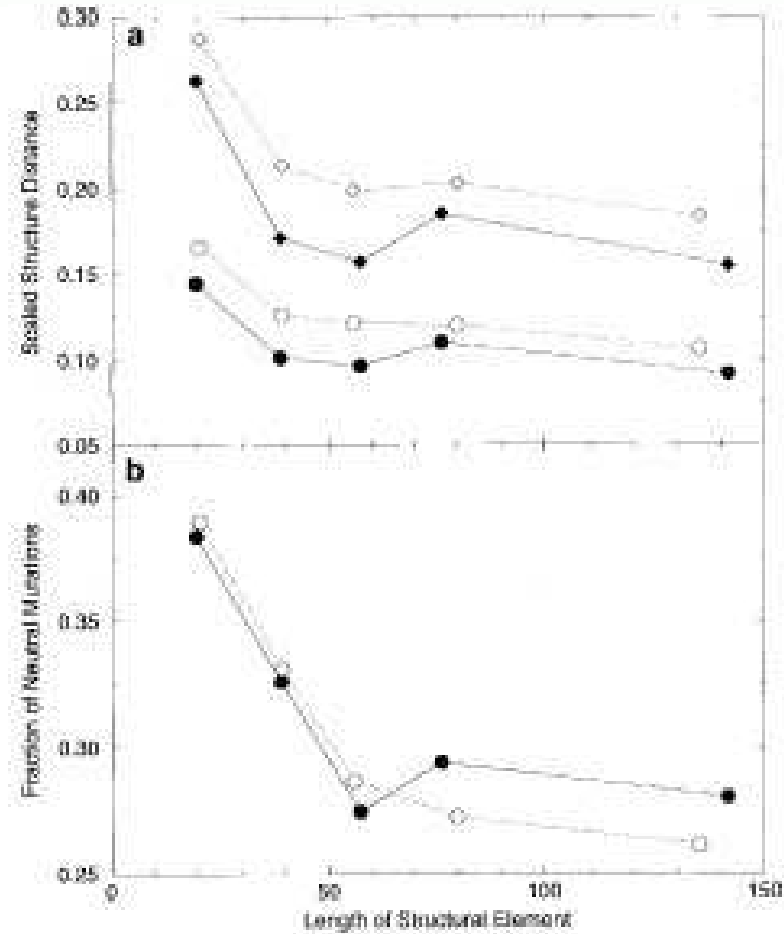
Viral RNA structures : Conserved

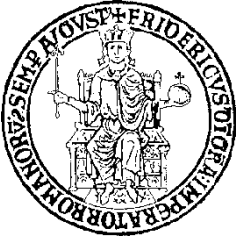


HIV sequence



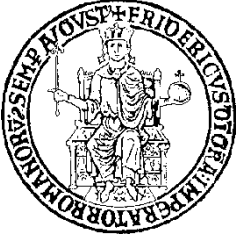
Robustness vs. length





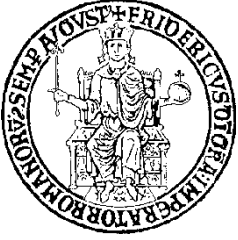
Conclusions

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Conclusions

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- ⑥ Mutational robustness entails non-optimality of binding sites (*fuzziness* of binding motifs)
- ⑥ There are indications that robustness of binding sites is optimized genome-wide (proportionality of affinity and pleiotropy)
- ⑥ Bacteria should exhibit a higher degree of variability in binding sites than eukaryotes but a smaller one than RNA viruses