1 The underlying philosophy of the model

The dynamical model is a schematic representation of the activity of genetic network. We have to discuss the assumptions the define the model from a biological point of view: the main criticism to a model is that its assumptions cannot be justified by the biological mechanisms. Our goal is to model the genetic activity related to a differentiation process of a cell: i.e. this activity is a stable long term activity whose stability is probably controlled by biochemical mechanisms (i.e. methylation processes), but for cancer cells the control dynamics is not so efficient allowing the evolution of different cell populations. Then we assume that this evolution is possible due to the competition of different genetic activities through dynamical mechanisms that can be triggered by the external environmental signals. In particular we assume:

- the long term genetic activity is determined by the presence of small genetic networks that have a stable active dynamical state;
- there exists an eternal control mechanism: the subnetworks have control nodes that prevent the arise of the active state in the subnetwork if there are set to the inactive state;
- once the active state has been established in a subnetwork it remains stable in time without any stimulus, except if an inhibitory stimulus change the state of control nodes;
- the stability and the controllability properties of a subnetwork depends from the existence of loops in the subnetwork: a loop may be related to the activation of metabolic cycles in the cell that define the cell behavior;
- each node of a subnetwork may represent the state of a gene that is connected and regulates the activation of other genes;
- in the cell differentiation mechanism is defined by the competition of different subnetworks that interact in a inhibitory way;
- the mutation mechanism change the connectivity of the network: we may distinguish between permanent changes and dynamical change (i.e. a connection may exist or non exist during time).

The complexity of the model is not a fundamental issue since we want to point out universal behaviors: first of all the existence of bistability or bifurcation phenomena for simple model and the definition of control parameters.

2 The mathematical model and related problems

Here we studied the model dynamics in different situations using mathematical methods. The main idea is understand the dynamics of the models to point out the universal properties that are robust and could explain the experimental data. The biological meaning of control parameters is a fundamental task to apply the model to predict the results of new experiments.
We consider a physical system that can be described by an weighted interaction network among nodes that can assume different dynamical states (in the case of

a gene network the states $\sigma \in [0,1]$ and we have models similar to spin models). The interaction structure is defined by signed adjacency matrix $A_{ij} \in [-1,0,1]$ where the sign refers to a cooperative or antagonist interaction between the connected nodes. In the simplest case, we introduce a stochastic dynamics using the probability $p_i(\sigma,t)$ that the node i is in the state σ (we assume $\sigma > 0$) at time t: in a deterministic approach $p_i(\sigma,t) = \delta(\sigma - \sigma(t))$ to denote that the node assume the state $\sigma = 1$ with probability one. The evolution of a deterministic model can be described by the equation

$$\sigma_i(t+1) = \Phi_i(\sigma(t)) = \Theta\left(\sum_j A_{ij}\sigma_j(t)\right) \tag{1}$$

where $\Theta(x) \in [0,1]$ is a threshold sigmoidal function (we assume $A_{ii} = 0$ to avoid self loops).
Remark: the dynamics is a information diffusion on the network. If we consider the linear system

$$\zeta_i(t+1) = \sum_j A_{ij}\zeta_j(t)$$

where ζ_i are non negative integers we have an equivalent dynamics since $\sigma_i = \Theta(\zeta_i)$ and it is possible to study the linear system to derive some properties of the initial system. For example the relaxation time to the solution $\sigma_i = 1 \ \forall \ i$ is for a given initial condition $\sigma_j^0 = \delta_{jk}$ is $t = n$ such that the matrix A^n has positive entries along the whole k-th column. This mens that for each node i there is a walk of length n from the initial node k to i.
We also assume a cause-effect relation so that A_{ij} is a directed graph. The deterministic model is a Hopfield network (each node has at least an input and an output link; the environment nodes has only output links) and one could study the equilibrium states and their stability. An equilibrium condition as follows is characterized as follows: for each i let

$$Q_i(t) = \sum_j A_{ij}\sigma_j(t)$$

then $Q_i > 0$ if $\sigma_i > 0$ and vice versa. Then $A_{ij} \geq 0$(i.e. A_{ij} is a connectivity matrix for a directed network) implies that the non trivial equilibrium is $\sigma_i = 1$: if $\sigma_k = 0$ for some $k \in K$ then we have

$$\sum_{j \notin K} A_{kj}\sigma_j = 0$$

so $A_{kj} = 0$ for all $j \notin K$ and the network is disconnected. Then we have the trivial solution $\sigma_i = 0$. For each equilibrium solution σ^* we have a stability basin

$$S_{\sigma^*} = \left\{\sigma \mid \lim_{t\to\infty} \sigma(t) = \sigma^*\right\}$$

If S_{σ^*} defined neighborhood of σ^* the solution is stable or if $S_{\sigma^*} = \{\sigma^*\}$ the solution completely unstable. The stability of the origin depends on the existence of a Ljapounov function: let introduce the network activity

$$\Sigma(t) = \sum_i \sigma_i(t) = \sum_i \Theta(Q_i(t-1)) \geq \Sigma(t-1)$$

since if each node has at least one input link, $A_{ij} = 1$ implies $\sigma_j(t-1) \Rightarrow \sigma_i(t) = 1$ and the activity cannot decrease. The solution $\sigma_i = 0$ is completely unstable. If there would exists an equilibrium solution with $\sigma_k = 0$ for some k then we define S_A the set of nodes s.t.

$$i \in S_A \quad \Rightarrow \quad \sigma_i = 0$$

(obviously $\sigma_k \in S_A$). Let $S_{\bar{A}}$ the complement of S_A, the network dynamics implies

$$0 = \sum_j A_{ij}\sigma_j = \sum_{j \notin S_A} A_{ij}\sigma_j = 0 \qquad \text{if} \quad i \in S_A$$

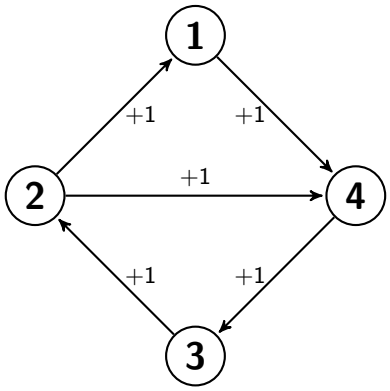
so that $A_{ij} = 0$ if $i \in S_A$ and $j \in S_{\bar{A}}$: i.e. there is not a cause-effect connection between $S_{\bar{A}}$ and S_A and the state $\sigma_i = 1$ for $i \in S_{\bar{A}}$ is an equilibrium state. Therefore we have as many equilibrium states as many partitions S_A and $S_{\bar{A}}$ there exist such that S_A triggers the activity of $S_{\bar{A}}$ but not vice versa. For any initial condition $\sigma_i(0) = \delta_{ik}$ the possible evolution are a periodic orbit or an equilibrium state: one can detect all the equilibrium conditions by σ^* by the condition

$$\sigma_i^* = 1 \quad \text{if} \quad \sigma_i(t) = 1 \quad \text{for some} \quad t \geq 0$$

The equilibrium states are a semigroup: let σ^a and σ^b two equilibrium states the

$$\sigma^a \cup \sigma^b = \sigma^c$$

is still an equilibrium. An example: if there exit a one directional loop γ in the network and there is no output link from γ to the remaining nodes of the network then $\sigma_i = 1$ for any $i \in \gamma$ is an equilibrium. If the loop is simple (each node has a one input link and one output link) the equilibrium is neutral since any change $\sigma_i = 1 \to \sigma_i = 0$ creates a periodic orbit (the total activity is constant). But if we we add a link to the loop then we get a stable solution since a single node can trigger the activity of two nodes and the equilibrium is an attractive stationary state (see figure). If a node is accidentally set to zero this anomaly propagates in the loop, until it reaches the node 4 where it is annihilated by the activity of the node (2). The average lifetime of a single perturbation is the average path length to propagate to the node (4) from the initial node (therefore it depends from the loop length or in case of presence of many loops, the average path length is computed considering independent loops).



The boolean network models the propagation of information. By studying the stability problem of the solution $\sigma_i = 1$ it is convenient to introduce the dual dynamics:

$$\sigma_i^c(t+1) = \Theta\left(\prod_{j\sim i} A_{ij}\sigma_j^c(t)\right) = \prod_{j\sim i} A_{ij}\sigma_j^c(t)$$

where $\sigma_i^c = 1 - \sigma_i$ is the dual state of the node and the product is restricted to the nodes connected to i ($A_{ij} \neq 0$): i.e. the node (4) takes the state $\sigma^c = 1$ only if both the nodes (1) and (2) in that state at previous time. This dynamics is valid for any configuration of the network and the state $\sigma^c = 1$ moves on the network until it reaches an absorbing state for which

$$\prod_{j\sim i} \sigma_j^c(t) = 0 \quad \forall\, i$$

For a given stable equilibrium σ^γ state associated to a loop γ any environmental perturbation that set to zero a activity of a node will destroy the equilibrium after a time equal to the number of the loop nodes minus one. For example in the figure there are two loops $((1) \to (2) \to (3) \to (4))$ and $((2) \to (4) \to (3))$ if we set to zero the node (4) after three iterations all the nodes will be in the zero state. The two loops are nit independent since one loops contains the other). On the contrary if we set to zero the node (1) one loop remains active. This remark allows to introduce the concept of control node: a node is a control node if its state is able to force the state of the whole network. The effect of a thermal bath could be introduced by assuming that the state of a node is defined as random variable that takes value $\sigma_i(t) = 1$ with probability $p_i(t)$ where

$$p_i(t+1) = \Theta_T\left(\sum_j A_{ij}\sigma_j(t)\right) \tag{2}$$

and $\Theta_T(x)$ is a logistic function

$$\Theta_T(x) = \frac{1}{2}\left(1 + \mathrm{tgh}(x/T - \epsilon)\right)$$

where ϵ measures to tendency of the network to be in the idle state when no stimulus is present. The logistic function is a generic sigmoidal function we do not expect that the specific form of $\Theta_T(x)$ is critical for the results.

Remark: since the values of x are quantized to integer in any case, if $\epsilon > T^{-1}$ the idle state is statistically attractive so ϵ could define a critical temperature for the network activation. We recover the deterministic dynamics for $T \to 0$. As a stochastic process we have a Markov process (since the realization of the variable $\sigma_i(t+1)$ depends only on the present state $\sigma_j(t)$ of the network. The dynamics (2) is a Markov field: the realization of the variable σ_i depends only from the present state of the network (and not from past states) and only from the states of the connected nodes $A_{ij} \neq 0$. The last condition (Markov field) means that the realizations of $\sigma_i(t)$ and $\sigma_j(t-1)$ are independent if the nodes are not connected. The transition probabilities depend from the state of the network and one derives the average dynamics

$$< \sigma_i > (t+1) = p_i(t+1) = \left\langle \Theta_T \left(\sum_j A_{ij} \sigma_j(t) \right) \right\rangle \simeq \Theta_T \left(\sum_j A_{ij} p_j(t) \right) \quad (3)$$

Then we have two possibilities: if the total average network activity tends to increase

$$\bar{\Sigma}(t+1) = \sum_i p_i(t+1) = \sum_i \Theta_T \left(\sum_j A_{ij} p_j(t) \right) > \bar{\Sigma}(t) \quad (4)$$

the equilibrium solution $\sigma_i = 1$ is attractive, on the contrary we have a an average tendency to decrease the network activity. The situation is illustrated in the fig. Remark: the mean field approximation apply when the $\Theta_T(x)$ can be approximated by a linear function locally: i.e. the fluctuations are small enough to approximate the function by a linear function in the whole fluctuation range. This is certainly not true when we have fat tail fluctuations.
Except for a small initial region, the condition (4) can be satisfied up to a critical value of the network activity Σ (if the temperature is not too big), so that the average activity tends to increase. But if the activity is below the critical value then the network activity tend to decrease and the stability of the solution $\sigma_i = 1$ is lost. A connected network tends to be more stable since the quantities $\sum_j A_{ij}\sigma_j$ increase. This picture is clearly an approximation since we neglect the fluctuation effects: if the fluctuations are big (this depends also on the connectivity matrix) we may have a fast transition between the two possible regime and a correction of the critical value. The critical vale is a consequence of the sigmoidal behavior of the $\Theta_T(x)$ function and its depends on the temperature and on the ϵ values. In presence of fluctuations and of two dynamical regimes (active and non active) we expect that the network activity may switch from one regime to another with a characteristic time scale (cfr. Kramer transition rate Theory). The transition may be triggered by large fluctuations that are both consequence of rare events (in such a case the probability should be exponentially small with

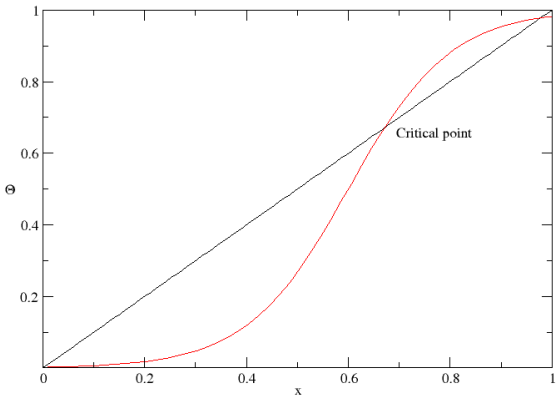


Fig. 1. Possible behavior for the condition (4); the units are arbitrary and scale with the network dimension.

respect the activity) but also depend on the network structure (the presence of hub nodes that can change the activity of many nodes amplifies the effect of small fluctuations (i.e. the change of the hub node state) and may introduce fat tail statistic in the fluctuation distribution). A second stochastic effect is related to the fluctuations of the connectivity due to environmental causes: the matrix $A_ij(t)$ is a stochastic process (so that its entries change their value according to a probability distribution). The simplest model can be formulated as follows: we assume that the nonzero entries $A_{ij}(t)$ assume value 1 with a given probability p (independent from the network state) each time step Δt (i.e. we are not simulating a parametric white noise, but a correlated random noise with a define correlation time scale Δt). Δt is the shortest evolution time scale for the system (we need a physical interpretation) and we set $\Delta t = 1$. The effect of a parametric noise is substantially different from the environmental noise and the evolution equation (1) reads

$$\sigma_i(t+1) = \Theta\left(\sum_j A_{ij}(t)\sigma_j(t)\right) \tag{5}$$

In such a case the average dynamics is not useful and the problem can be studied by the representative dynamics

$$\zeta_i(t+1) = \sum_j A_{ij}(t)\zeta_j(t) \qquad \Rightarrow \qquad \zeta(t) = \prod_{k=1}^{t} A(k)\zeta(0)$$

where the solution is the product of random matrices (there are results on the spectral properties). From a biological point of view means that the interaction of genes depends also by external factors.

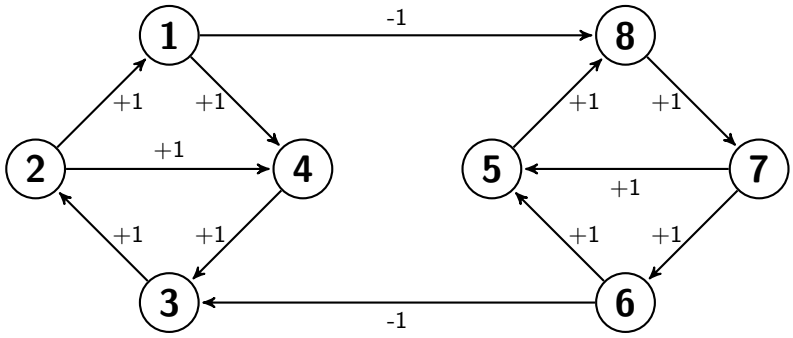
One could say that the network is active if a certain condition is satisfied (for example the average total activity should overcome a given threshold) so that fluctuation may introduce the existence of non active states. Problems for a single network: starting from a loops with a fixed dimension adds randomly links to stabilize the equilibrium solutions (existence of sub-loops) and study the robustness of the solution and the recovery times in relation with the connectivity matrix; adding the temperature, study the existence of critical value for the appearance of the equilibrium solution; the thermodynamic limit. The effect of the environmental noise has to be justified from a biological point of view by relating it to the individual variability of the cell phenotypes in an homogeneous population.

In the stochastic models one should also consider the problem that the connectivity matrix is not fixed (for example we have a ensemble of admissible matrices or the existence of the links is a random event). In such a case we have a stochastic dynamics

$$\sigma_i(t+1) = \Theta\left(\sum_j A_{ij}(t)\sigma_j(t)\right) \tag{6}$$

where $A_{ij}(t)$ is a random process with value $\in \{0,1\}$ maintaining some average properties of the connectivity; this is an alternative to the environmental noise (parametric noise). This model simulates the fact that the activation of a link may depends on random events (i.e. not only from the existence of the link) so that a genetic network is indeed a stochastic network. In principle any realization of the connectivity matrix A_{ij} has an equilibrium $\sigma_i = 1$ but the robustness of equilibrium can be influenced by the fluctuations. Problem: if the robustness of the equilibrium with respect to the external perturbations (i.e. an external signal on a node) depends on the spectral properties of the connectivity matrix (to be studied) then it is possible to study the spectral properties of random connectivity matrices (see Catanzaro thesis) and develop a control theory for the network.

Let us consider the existence of competitive networks (see figure) that are linked by inhibitory links: if the first network is in an exited state the second network should be completely switched off for a stable equilibrium.



If we start with all the node states set to one we create a frustrated situation, otherwise the network choose one of the two possible stable states. In such a case the presence of an environmental noise could induce the transition to one state to another (to be studied). An external forcing breaks the symmetry.
We expect a transition phase as a function of the temperature: increasing the temperature the node states tends to be independent, but under a threshold the system should choose a stationary state.
The system can be generalized to consider the interactions of different cooperative networks (possibly with different internal structure) that are connected by inhibitory links (in the case of connection with excitatory links we join the subnetwork in a single one). We can introduce a metadynamics where $\nu_k(t)$ is the state of the k subnetwork and we have a relation

$$\nu_k(t+\Delta t) - \nu_k(t) = \phi(\nu_k(t)) - \gamma\left(H_{kj}\nu_j(t)\right) \tag{7}$$

where $H_{hk} \geq 0$ is an inhibitory connectivity matrix. $\phi(\nu_k(t))$ describes the tendency of the sub-network to increase its activity and γ the average decreasing of the activity due to the presence of other sub-networks. This is an effective equation: ν_k should describe the network activity (i.e. it could be the time-average activity of the nodes assuming that the network could be considered in a stationary state). Indeed the evolution time scale Δt could be assumed $\Delta t \gg 1$ so that the subnetwork states are relaxed to a stationary states. The structure of attraction basins of the stable states could be related to a potential in the state space if

$$\nu_k(t+1) - \nu_k(t) = -\frac{\partial}{\partial \nu_k}\left[\frac{\gamma}{2}\sum_{ij}\nu_i H_{ij}\nu_j + \sum_j V(\nu_j)\right]$$

where

$$\phi(\nu) = -\frac{\partial V}{\partial \nu}$$

Since $\phi(\nu) \geq 0$ $V(\nu)$ is increasing. Then we introduce the energy

$$E = \frac{\gamma}{2}\sum_{ij}\nu_i H_{ij}\nu_j + \sum_j V(\nu_j)$$

and the equilibrium are the critical points of the energy. Moreover

$$E(t+1)-E(t) \simeq (\nu_k(t+1)-\nu_k(t))\frac{\partial}{\partial \nu_k}\left[\frac{\gamma}{2}\sum_{ij}\nu_i(t)H_{ij}\nu_j(t)+\sum_j V(\nu_j(t))\right]$$
$$= -\frac{1}{2}\frac{\partial}{\partial \nu_k}\left[\frac{\gamma}{2}\sum_{ij}\nu_i(t)H_{ij}\nu_j(t)+\sum_j V(\nu_j(t))\right]^2$$

Therefore the energy is a Ljapounov function and the system equilibria are defined by the critical points of the Energy function corresponding to local minima and maxima.
Remark: the existence of the Energy implies that H_{ij} is symmetric negative defined

$$\frac{\partial^2 E}{\partial \nu_j \partial \nu_i} = \frac{\partial^2 E}{\partial \nu_i \partial \nu_j}$$

The stochastic effect has to be introduce but it is possible a thermodynamics approach and a thermodynamics equilibrium exists according to the Maxwell-Boltzmann distribution and the detailed balance condition. This means that the whole network does not satisfy this condition, but the metadynamic network realized a reversible Markov process. The existence of a thermodynamic equilibrium allows to use Maximal Entropy Principle and the Maxwell Boltzmann distribution when we introduce a thermal bath.
We are interested in networks with many different equilibria each one related to exited state of subnetworks (or a combination of subnetworks), in the effect of a thermal noise and in the effect of external forcing. The external we introduce in the network boundary nodes whose state is defined by a given external signal $\sigma_b(t)$ (possible a stochastic process) then the network dynamics reads

$$\sigma_i(t+1) = \Theta\left(\sum_j A_{ij}\sigma_j(t)+\sum_b A_{ib}\sigma_b(t)\right)$$

where A_{ib} is the link between the environmental node b and the node i. It is possible to introduce a probabilistic description of the evolution of the probability that the network is in the state σ' at time $t+1$ according to

$$p(\sigma', t+1) = \sum_\sigma \pi(\sigma', \sigma)p(\sigma, t) \tag{8}$$

where

$$\pi(\sigma'|\sigma) = E\left(\delta_{\sigma', \Phi_{\sigma_b}(\sigma)}\right)$$

and the expectation value is computed on the realization of the input noise. $\pi(\sigma'|\sigma)$ is the transition rate per unit time (the continuous limit could be considered). Let σ_{eq} stable equilibrium state the effect of external random perturbations could be to move the network state in a neighborhood of the equilibrium

solution or it could induce a transition to other equilibrium basin attractions so that the dynamics starts to perform an intermittence behavior. In such a case the relevant quantities are the residence times in the different basins that can be associated to metastable states.
We introduce the stochasticity in the system assuming that the adjacency matrix is not known: i.e. A_{ij} is a extracted from an ensemble of random matrices. As the result of an experimental one could assume that each entry A_{ij} is a dichotomous random variable with probability p_{ij} to get the value ± 1 (i.e. the link is active). The value $p_{ij} = 0$ is admitted so that the corresponding link it always inactive. The problems are:

1. Classifying the equilibrium states in relation to their robustness with respect the changes in the adjacency matrix;
2. Understanding the representativity of the average dynamics: i.e. substituting the adjacency matrix with an average matrix one highlights the dynamical properties that are correctly described by the average system
3. Pointing out the existence of bifurcation phenomena so that it is possible to divide the ensemble in different communities with similar dynamical behaviors.