Core percolation and onset of complexity in Boolean networks

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The determination and classification of fixed points of large Boolean networks is addressed in terms of constraint satisfaction problem. We develop a general simplification scheme that, removing all those variables and functions belonging to trivial logical cascades, returns the computational core of the network. The onset of an easy-to-complex regulatory phase is introduced as a function of the parameters of the model, identifying both theoretically and algorithmically the relevant regulatory variables.

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Introduction — Boolean Networks (BN) are dynamical models originally introduced by S. Kauffman in the late 60s [1]. Since Kauffman’s seminal work, BN have been used as abstract modeling schemes in many different fields, including cell differentiation, immune response, evolution and gene-regulatory networks (for an introductory review see [2] and references therein). In recent days, BN have received renewed attention as a powerful scheme of data analysis and modeling of high-throughput genome and proteome experiments [3].

The central issue of previous research was the description and classification of different BN attractor types under deterministic parallel update dynamics [3,4,6]. Special attention was dedicated to so-called critical BN [6], situated at the transition between ordered and chaotic dynamics. We follow a complementary approach: Our main goal here is to study the statistical properties of fixed points of the dynamics of general BN. We first reformulate the original dynamical problem into a constraint satisfaction problem, and then we study it with techniques borrowed from statistical mechanics of disordered systems, cf. [7]. As a result, we are able to classify different types of fixed-point organization, and to identify the set of relevant regulatory variables.

The model — In Kauffman networks, the expression of one gene is modeled by a Boolean variable: the expression level of a regulated gene $a$ is determined by the expression levels of the transcription factors $a_1, ..., a_K$:

$$x_a = F_a(x_{a_1}, ..., x_{a_K})$$

with $a \in A \subset \{1, ..., N\}$ running over all regulated genes.

The central question of this work is: are methods from the statistical mechanics of disordered systems able to capture number $\mathcal{N}_{fp}$ and organization of stationary points of this network, i.e. vectors $\vec{x}$ fulfilling simultaneously all equations of type (1) with $a \in A$? The main step here is to write them as zero-energy ground states of a cost function, or Hamiltonian, defined as

$$\mathcal{H}(\vec{x}) = \sum_{a \in A} x_a \oplus F_a(x_{a_1}, ..., x_{a_K}) .$$

The symbol $\oplus$ stands for the logical XOR operation, i.e. each cost term contributes zero to the sum if Eq. (1) is fulfilled, and one otherwise. The Hamiltonian $\mathcal{H}(\vec{x})$ thus counts the number of unsatisfied Boolean equations. This embeds the problem of finding fixed points into the class of constraint-satisfaction problems, which have been recently studied extensively from the point of view of statistical physics [7], using various techniques from spin-glass physics, in particular the cavity method [8]. Note that a related approach based on topological properties of the BN was independently proposed in [10].

FIG. 1: Example for a small Boolean network, circles symbolize variables, squares Boolean functions. $x_1$ is an example for an external input variable, $x_4$ for a functional variable, whereas $x_3$ stands for a regulatory variable, see below.
In this work we concentrate on the interplay of different types of functions (represented by $F_a$) for given probability measure of the topology of the network (represented by the $a, a_1, \ldots, a_K$). For the moment we restrict the analysis to the case of only $K = 2$ inputs. Extensions will be discussed at the end of this Letter.

Let us assume that the network is random, i.e. its topology is described by a random directed hyper-graph where the triples $(a, a_1, a_2)$ are randomly chosen with the following conditions: (i) A function is regulated by at most one function, i.e. any two distinct triples $(a, a_1, a_2)$ and $(b, b_1, b_2)$ fulfill automatically $a \neq b$. (ii) There are $M = \alpha N$ of these triples. Condition (i) restricts the fraction $\alpha = M/N$ to the interval $[0, 1]$. We can distinguish three relevant types of variables as displayed in Fig. 2.

FIG. 2: External input variables: There are $N - M$ variables which are not regulated by any function. They represent external inputs to the network. Regulatory variables are all those variables which are regulated and regulate. Functional variables: There are $e^{-2\alpha}N$ variables which do not regulate any other function.

We now have to specify the functions acting on top of the random topology. There are $2^{2^K} = 16$ Boolean functions, which can be grouped into 4 classes [11]: (i) The two constant functions. (ii) Four functions depending only on one of the two inputs, i.e. $x_1, \overline{x}_1, x_2, \overline{x}_2$. (iii) AND-OR class: There are eight functions, which are given by the logical AND or OR of the two input variables, or of their negations. These functions are canalizing in the sense that, e.g., if $F(x_1, x_2) = x_1 \land x_2$ and the value of $x_1$ is set to zero, the output is fixed to zero independently of the value of $x_2$. It is said that $x_1$ is a canalizing variable of $F$ with the canalizing value zero. (iv) XOR class: The last two functions are the XOR of the inputs, and its negation. They are not canalizing: Whatever input is changed, the output changes, too.

Here we are only interested in true $K = 2$ functions, i.e. in the AND-OR class and the XOR class, but the extension of our analysis to the whole spectrum of $K = 2$-functions is a minor technical point. The organization of fixed points does not depend on the relative appearance of different functions within each class, but only on the relative appearance of classes. We therefore require $xM$ functions to be in the XOR class, and the remaining $(1 - x)M$ functions to be of AND-OR type, with $0 \leq x \leq 1$ being a free model parameter. In this sense, for $K = 2$, the network ensemble is completely defined by $\alpha$ and $x$.

Computational core — Many variables can be fixed following logical cascades starting in external variables, and the corresponding Boolean equations become fulfilled. Other functions can be satisfied trivially because they include variables being otherwise unconstrained (see below). However, a certain set of equations (depending both on the BN and the configuration of external variables), cannot be satisfied on the basis of such simple local arguments. We define the computational core (CC) as the maximal subnetwork formed by these functions.

The first process to be considered in this context is propagation of external regulation (PER). If both inputs of a function are fixed externally, or if a canalizing input is fixed to its canalizing value, also the output is directly fixed externally. This process can be propagated, exploiting all direct logical implications of the configuration of the external input variables: all the functions which are trivially satisfied by this process can be removed. The probability that a function survives is

$$\pi_{PER} = 1 - (1 - \alpha \pi_{PER})^2 - (1 - x)(1 - \alpha \pi_{PER}) \alpha \pi_{PER},$$

i.e., neither both input variables are fixed by PER nor, in the case of a canalizing function, a canalizing variable is fixed by PER to its canalizing value. For small $\alpha$, this equation has only the trivial solution $\pi_{PER} = 0$, i.e. all variables are completely fixed by PER with probability one. At $\alpha_{PER}(x) = 1/(1 + x)$, a new solution appears continuously. Above this point, a finite fraction of all functions survives the PER decimation process, forming the PER core. The core percolation point moves toward one in the limit $x \to 0$: in a pure AND-OR network an extensive PER core never exists due to the canalizing character of the functions. PER is closely related to the percolation of information discussed in [2], which studies the parallel evolution of two slightly distinct configurations. Identifying external variables in our model with constant functions in Kauffman’s original formulation, the phase transition line becomes exactly the location of critical BN. The region left to the PER transition line corresponds to ordered, while the region on its right corresponds to chaotic networks under parallel dynamics [12].

The second process in determining the core is the leaf-removal procedure (LR). A leaf is a variable which has degree one, i.e. a variable which is either functional (regulated but not regulating) or which is not regulated, and acts exactly on one other variable. A regulated leaf corresponds to a Boolean equation which can be trivially fulfilled, the same is true if both input variables of a (non-constant) function are leaves. In the XOR case, even a simple input leaf allows for satisfying the Boolean equation independently of the values of the other input and the output [12]. None of these cases induces complex behavior based on frustration - meaning in this context that the fixed points of the Boolean network are easily determined - and can be eliminated from the network. This process may induce new leaves, and can be iterated until no removable function is left. The survival probability of
a randomly selected Boolean function reads now:

\[ \pi_{LR} = x(1 - t_{\text{out}})(1 - t_{\text{in}})^2 + (1 - x)(1 - t_{\text{out}})(1 - t_{\text{in}}^2) \]

It depends still on the probability that a variable at a certain point in this process becomes a regulated \( t_{\text{out}} \) or regulating \( t_{\text{in}} \) leaf, which can be determined self-consistently using an iterative argument:

\[ t_{\text{out}} = \exp\{-2\alpha (1 - t_{\text{out}})(1 - x t_{\text{in}})\} \]

\[ t_{\text{in}} = (1 - \alpha x (1 - t_{\text{in}}^2) - \alpha (1 - x)(1 - t_{\text{in}}^2)) t_{\text{out}} \]

There is always the solution \( \pi_{LR} = 0 \) corresponding to complete removal of all functions by LR. At some \( x \)-dependent \( \alpha_{LR}(x) \), a second solution appears discontinuously, and the LR core size jumps to a finite fraction of the complete network. The line \( \alpha_{LR}(x) \) is monotonously growing in \( x \), going from 1/2 for the pure AND-OR case to 0.8839 for the pure XOR case.

Both procedures can be combined in order to determine the computational core of the Boolean network under one external condition: First LR is applied, and then PER is needed to see which variables on the LR core are fixed via propagation of external regulation. The problem can still be solved analytically. Here we give only the results, technical details will be presented elsewhere. For \( x > 0.636 \), the emergence of the CC is discontinuous and appears exactly at the point \( \alpha_{LR}(x) \). The LR core thus contains immediately an extensive computational core. The height of the discontinuity decreases, however, if we approach \( x \approx 0.636 \) from above, and we find a tricritical point in \( (x, \alpha) = (0.636, 0.785) \). Below this point, the CC emerges only after the LR percolation transition, and the transition is continuous. The results are summarized in the phase diagram of Fig. 3.

**Clustering of solutions and complexity** — What does the existence of an extensive CC imply in the structure and organization of fixed points? This question can be answered by applying the zero-temperature cavity method of statistical mechanics to the Hamiltonian \( H(\vec{x}) \) defined in Eq. 2; technical details will be presented in a longer publication. The first result concerns the solution entropy which is found to be

\[ s = \frac{1}{N} \log(N_{fp}) = (1 - \alpha) \log(2) \]

with the over-bar denoting the average over the random network ensemble with fixed \( \alpha \) and \( x \) (with, in fact, no dependence on \( x \)). This implies that fixed points satisfying all Boolean functions exist for all \( \alpha < 1 \) and all \( x \). The line \( \alpha = 1 \) can be considered as the SAT/UNSAT transition line. The entropy value shows that each configuration of the \( (1 - \alpha)N \) external input variables leads on average to one stationary point, i.e. the state of all internal and functional variables is mainly determined by the external conditions, even in the case of a PER core, where this fixing is not just a simple logical implication. Even if the entropy is analytic in all the phase diagram, the organization of the fixed points in configuration space undergoes a clustering transition at some \( \alpha_d(x) \), see Fig. 4. Below it, all solutions are collected in one cluster: any pair of them can be connected by a path via other solutions, where in each step only a finite number of variables can be changed. The solution space is technically named replica symmetric (RS). At the clustering transition, the replica symmetry breaks spontaneously (RSB): an exponential number of macroscopically separated clusters of fixed points appears. Their number, or more precisely its normalized logarithm, is called complexity. It jumps discontinuously at \( \alpha_d(x) \), as can be seen in Fig. 4. The clustered phase sets on in general well inside the region where a CC exist. Exceptions are the extreme points \( x = 0 \) (neither core nor clustering appear at any \( \alpha < 1 \)) and \( x = 1 \) (both transition points coincide).

**Beyond** \( K = 2 \) — The case of Boolean functions depending on exactly \( K = 2 \) can be generalized. However, the number of Boolean functions of \( K \) variables diverges as \( 2^2^K \). For \( K = 3 \), the 256 functions can still be classified completely: there are 14 classes, 4 of them are the classes already discussed in the context of \( K = 2 \), and only 10 lead to actual \( K = 3 \) functions. For \( K \geq 4 \), non-exhaustive numerical checks were done. The main results are: (i) The minimum of the clustering point \( \alpha_d \) over all combinations of functional classes is always given by the pure \( K \)-XOR class (the only completely analytically accessible). We find \( \alpha_d(K = 3) = 0.782, 0.699 \) for \( K = 3 \), and \( \alpha_d(K = 4) = \ln(K + \ln K + \ln \ln K + ...)/K \) for \( K \gg 1 \). Note that the range of the complex phase becomes larger...
with $K$. (ii) We conjecture that, for fixed $K$, the only class not leading to clustering is the pure $K$-AND-OR one. This has been checked explicitly for $K = 3$, and non-exhaustively also for $K = 4$. This class becomes, however, both combinatorially and biologically less important for higher $K$, where threshold-like functions are expected to be more relevant.

**Conclusion and outlook** — We have analyzed the organization of fixed points in random Boolean networks identifying the sudden emergence of a computational core, whose existence is a necessary (but not sufficient) condition for a globally complex phase where all fixed points are organized in an exponential number of macroscopically separated clusters. This phenomenon is found to be robust with respect to the choice of the Boolean functions, and missing only in networks completely made of AND and OR functions. In addition, the size of the complex regulatory phase grows if a higher number $K$ of inputs to the Boolean functions is allowed.

**Open Questions** — Our analysis is based on ground states of a Hamiltonian counting the number of violated functions, not directly related to any overlying biologically motivated dynamics. The *dynamical accessibility* of fixed points remains an open challenge which will be addressed in a future project. Moreover, noise sources present in real cases may force the network to quasi-stationary points close to the fixed ones (some regulation might not always function properly, without effecting the overall state of a cell). In this view, the study of the organization and accessibility of meta-stable states in the region of low complexity and in the case of fixed external inputs, together with their relation with quasi-stationary points might be very relevant.

A second interesting issue is moving from average case analysis to real networks samples. Our method can be directly implemented as a *message passing algorithm on single network instances*, and not only on random ensembles as discussed in this work. However, very few genome-wide data are so far available. In particular, for multi-cellular organisms only small functional modules for well-described functions are known, and large-scale networks known for yeast [14] and *E. coli* [15] contain only the topological structure, not an extensive description of the regulatory functions. It is thus highly interesting to infer gene regulation networks from experimentally easily accessible high-throughput experiments, as e.g. given by genome-wide expression profiles [10]. This inverse problem could be treated with tools similar to the ones used in the present analysis.

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