Nonlinear Development of Bacterial Colony Modeled with Cellular Automata and Agent Objects

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Abstract

Collective dynamical behavior of simple organisms is a very fascinating and important field of study especially in the age of rapid development of nano- and bio- technology. Despite a number of different numerical techniques existing for modeling the uptake of the nutrients, metabolism, maintenance, cell division and growth of bacteria population, none of them can be treated as a universal one. This is because the complex behavior of simple organisms must be studied in different aspects covering totally different spatio-temporal scales. Most of numerical models employ both well known discrete techniques such as diffusion-(reaction)-limited algorithms, cellular automata, Monte-Carlo and continuum approaches. In a new model presented here we have combined two techniques. The first one – agent based – has been used for modeling the behavior of an individual bacterium. The agent defines generic features of the bacterium and the ways it interacts (communicates) with the environment and with the neighboring bacteria. The cellular automata is used for modeling the bacterial environment and represent communication layer for the agents, while a fixed two-dimensional grid defines the living space. Despite the entire system is treated as a system with unbounded resources, the resources are limited locally due to congested environment. The growth of the bacterial colony depends on the amount of free space in the closest neighborhood of individuals, which is required for reproduction, and on the availability of nutrients. We have matched the parameters of the model to demonstrate various growth structures developed by bacteria populations. We show that the patterns generated by the bacteria due to their collective behavior reflect the dynamical vitality of population and its fitness factor. We observe that the strongest populations self-organize in rod-like structures, which are reproduced in experimental light microscopy images characteristic for some bioflims and anthrax bacterial colonies.

Keywords: numerical modeling, bacteria evolution, anthrax, cellular automata, agent systems

Submitted to International Journal of Modern Physics C, February 2003

1 Introduction

The anthrax bacteria (*bacillus anthracis*) became well known this past year since it was used as a serious terrorist weapon (see e.g. [Todar; Turnbull; web pages]). To find out the ways for protecting populated area against this lethal disaster, we have to study the behavior of bacterial population in various types of environments (e.g., nourished or starving, liquid or solid) and a broad diversity of physical and chemical conditions, such as: very high and very low temperature and pressure, excess or lack of oxygen or acids. Some of these systems cannot be modeled in laboratories due to, e.g., high risk of loosing control on bacteria propagation, which may result in dangerous infections. The only issue is numerical modeling and simulation. The numerical modeling of dangerous bacteria and viruses focus on their propagation in a variety of physical conditions and environments (see e.g., [Gonpot et., al., 2000]), on morphology of microorganisms (e.g. [Kozlovsky et. al, 1999; Hermanowicz, 2001]) and on the collective behavior of the entire bacterial colony ([Gallas et. al., 1992; Cohen et. al., 1999; Ben-Jacob et. al., 2000]).

A single bacterium can be treated as a complex molecule, which interacts with environment, communicates with other individuals, replicates and undergoes evolution and mutation. The collective behavior of bacterial colony - reflected in a variety of patterns they create (see [Microbial Word, web page]) - indicates its fitness and adaptation abilities [Shapiro, 1995]. Pattern formation in microorganisms can be viewed as the result of the exchange of information between the microscopic objects (bacterium) and the macroscopic ensembles (the population). In result of employing different communication layers the bacteria population can be composed of smaller clusters of different shape. For example, *bacilli* cell may occur singly or form chains of cells; *cocci* may form chains (*streptococci*) (see Fig.1a) or grape-like clusters (*staphylococci*), the rod-like pattern shown in Fig.1b is the most characteristic pattern formed by the anthrax colony [Todar; Turnbull; web pages] and some biofilm structures [Gonpot et. al. 2000]. The anthrax bacteria develop straight rods and chains with arcs and branching structures. Conversely, the creation of singular resting spores is one of important adaptation feature of some viruses and bacteria including anthrax. The spore is a hibernated form of bacterium, which enables its survival in a hostile environment. The spore cannot nourish and reproduce. De-hibernated spore adapts in friendly conditions and undergoes fast evolution.

The bacterial chains and spores produce large colonies such as biofilms The biofilms represent a particular type of bacterial population [Gonpot et. al. 2000; Hermanowicz 2001]. They are composed of micro-organisms adhering to surfaces in which sufficient moisture is available. The biofilms are present in living organisms (both plants and animals) as well as environmental surfaces such as rocks

We propose here a new discrete model of bacterial colony, which is focused on modeling the evolution of populations consisting of rod-like clusters. We combine two dimensional cellular automata with the agent techniques (see e.g. [Wooldridge 2002]). Such the hybridized model can decrease large computational and memory complexity, which is typically required by agent–based systems. Additionally, the

model is more flexible then typical cellular automata by enabling modifications and control of individual features of a single bacterium. By using this model we address the following problems:

1. How microscopic synthetic rules influence the creation of the rod-like pattern?
2. Which important information is hidden behind this particular pattern?
3. Which way does the congesting environment influence on the entire bacterial colony?

Consequently, we try to match the microscopic rules, which are responsible for specific macroscopic patterns created by the bacteria colonies and correlate them with fitness factors of the population.

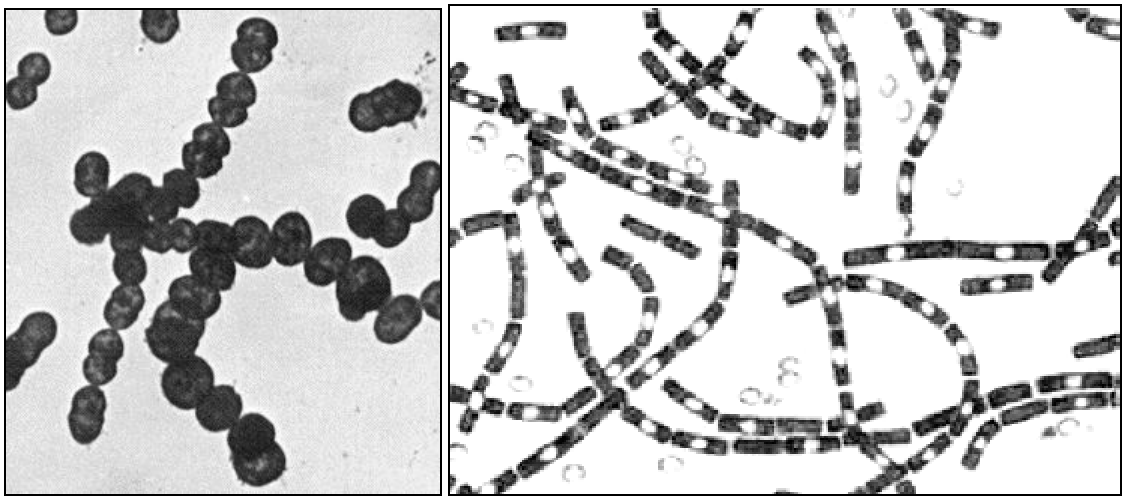

Figure 1 Typical chains of a) dividing streptococci, b) *Bacillus anthracis* – experimental microscopy image. The anthrax rod sizes are 1 - 1.2μm in width x 3 - 5μm in length

In the first section we discuss briefly the continuum and discrete models used for modeling bacterial colonies. We then introduce our algorithm and its principal assumptions. The results of simulations are demonstrated in the following section. We present some growth patterns, which are dependent on the local rules, and reproduce realistic shapes of colonies produced by amorphous or rod-like clusters. This has been achieved by including a realistic description of the bacteria behavior and the ways in which it absorbs food in a friendly environment and reproduces. Finally, we discuss the conclusions and summarize our findings.

2 Continuum and discrete models in modeling bacterial populations

In the modeling of the collective behavior of microorganisms, relevant space scales of centimeters might be represented by continuum mechanics in the form of the Navier-Stokes equations. The discrete models are related to the smaller scales (hundreds of micrometers), which size depends on the volume of constituents such as viruses, bacteria, polysaccharids etc. Both continuum and discrete models have to be used for

covering multiresolution aspects of biological systems [Cohen et. al. 1990], i.e., their spatio-temporal diversity starting from molecular structures of a single organism, through its genetic features, metabolism, interaction with an environment, population growth, and finally, complex macroscopic behavior. For example, the modeling of biofilm explores the relationship between pattern formation process and stochastic (and chaotic) micro-events on the organization of microbial communities [Wanner and Gujer, 1987, Eberl et. al. 2000]. There are many categories of both continuum and discrete models used for biofilm modeling.

The classical model of Wanner and Gujer [Wanner and Gujer, 1986], which mimics the growth of the biofilm's entities with differential equations, is a good example of the continuum approach. In this rather general concept it is assumed that smooth functions, which result from averaging, describe the bioflms' relevant aspects. Another example represents the problem of nutrient consumption in biofilms, which is governed by a convection-diffusion transport equation in the liquid environment and by diffusion and reaction in the biofilm itself [Picioreanu, 1999; Eberl et. al. 2000]. In the continuum model presented in [Cohen and Ben-Jacob, 2001] the bacteria are characterized by a density field with non-linear diffusion and a complex scalar field representing bacteria orientation.

The discrete models can be used most appropriately for modeling phenomena in which group behavior comes from microscopic interactions between independent objects and the objects and environment. These interactions can be expressed by analytic formula coming from basic principles or synthetic set of rules. The collective motion such as fingering, convection rolls and other micro-hydrodynamic instabilities involving by two ensembles of particles penetrating one with another [Dzwinel et. al. 2000; Dzwinel and Yuen, 2001; Dzwinel et. al., 2002], can be an example of the former type of interactions. Growth of biofilm differs from growth of a conventional thin-film because the specimens have a special shape and size. They are much larger than individual atoms or molecules and move slowly, or not at all, over the substrate surface compared to the diffusion of atoms on substrates. Despite of this difference, the evolution of multiple objects, which local rules cannot be described within known physical systems and classical mathematical formalism, still can be modeled by using similar tools as those employed for solving N-body problems in physics. For example, diffusion limited aggregation (DLA), diffusion-reaction growth and BAM (ballistic aggregation model) are discrete, random walker based models that are being used for modeling spatio-temporal patterns observed in creation of e.g., colloidal aggregates [Meaken, 1988, 1999]. As shown in [Matsushita and Fujikawa 1990; Schindler and Rovensky, 1994; Schindler and Rataj, 1992] the same methods can be also successfully employed in modeling the morphology of bacterial colony.

Many of biological films are impossible to realize both by using differential equations and discrete models. Therefore, heterogeneous continuum-discrete models were devised in [Picioreanu et. al. 1999; Gonpot et. al. 2000] in which reaction – diffusion limited aggregation models and continuum growth equations are combined together to mimic biofilm evolution. For example, in [Lacasta et. al. 2000], the authors present the models based on reaction and diffusion limited growth techniques for simulating

the evolution of *Bacillus Subtilis*. In [Ben-Jacob et. al. 2000, Cohen et. al. 1999, Golding et. al. 1999, Kozlovsky et. al. 1999] the discrete and continuum methods were combined for reproducing branching and chiral growth of starving colonies for the various types of bacteria (see [Bacterial Cybernetics Group, web page]).

The macroscopic models of growth involving scales of 1-5 milimeters can be also modeled within a unique discrete numerical paradigm such as cellular automata. Cellular automata (CA) are well-known systems consisting of a large set of primitive discrete-state elements interacting via a given set of local rules [Pires et. al., 1990; Stauffer, 1991; Rothman and Zaleski, 1994; Chopard and Droz, 1998, Wolfram 2002]. The CA rules act locally but reveals in a global behavior of the entire system. This kind of global behavior typically cannot be predicted easily from the microscopic rules. There are many different types of CA-based models used for studying the evolution of living organisms [Ermentrout and Edlestein-Keshet, 1993]. We can recognize four main classes of CAs. They are as follows:

1. Grid based CA – identical c_i automata are fixed in the nodes of a regular grid. The state of each automaton is synchronically updated according to a prescribed set of rules involving the states of the nearest neighbors of c_i.
2. Deterministic Eulerian automata – they mimic the solution of partial differential equations and can model oscillations, e.g., in excitable media (e.g. Belousov-Zhabotinsky reaction), cardiac function (see in [Delmore and Mazoyer, 1998] and Lotka-Volterra predator-prey dynamics.
3. Lattice gas models – the particles move on a discrete grid and collide in its nodes [Frish, 1987; Rothman and Zaleski, 1994; Chopard and Droz, 1998].
4. Solidifications models [Kremeyer, 1998] – particles are bounded and cannot move. The models simulate phase transition phenomena (e.g. solidification [Kassner et. al. 1998] and phase separation [Chopard and Droz, 1998]).

The cellular automata do not support flexible mechanism for definition, diversification and control of individuals expanding on the lattice. The agent systems (see e.g., [Wooldridge, 2002]) have built-in such the mechanism. The agents are defined as independent processes, which can interact, exchange information, die and can be modified by environment or other agents. There are many publications describing agent–based models (see e.g. [D'Inverno and Luck 2001]) and their implementations for modeling living organisms. For example, the system called Swarm accomplished in the Santa Fe Institute uses collections of concurrently interacting agents for numerical modeling of a variety of complex systems. As shown in [Kreft et. al. 1998], this framework can be used also for constructing a generic simulator for bacterial colony growth.

One of the serious drawbacks of agent objects approach is a large memory load they required. In the following section we show that by combining cellular automata with agent objects we can decrease computational and memory requirements of agent–based systems, simultaneously increasing flexibility of a typical cellular automata model.

3 The model of bacterial colony

In our model, bacteria are treated as independent agents, which interact with the environment and reproduce accordingly to the set of rules. The rules reflect two particular goals: growth and multiply. We define the agent as an object, which perceives its neighborhood by using sensors and acts autonomously in an environment influencing on it by means of effectors. In software independent agent systems the agents – implemented as a special kind of object - perform a similar code and are aware of the existence of the others agents. The agents communicate (interact) with each other by means of communication layers defined as the properties of the environment in which they operate.

We model the environment as a two-dimensional lattice of cellular automata, which represents the medium with unbounded resources, that is, free space, nutrients and other grow factors are supplied to the system without limit. However, the resources are limited locally due to congested environment, which restricts both the local space for reproduction and amount of nutrients in the densest places on the lattice. The resources are also limited due to finite size of the computational box. However, we stop our simulations at the moment when simulated bacterial colony reaches the borders of the box. We assume additionally that:

1. The purpose of bacteria existence is to grow. Growth is an orderly increase in the quantity of bacteria constituents. It depends upon the ability of the bacterium to form new protoplasm from nutrients available in the environment. In most bacteria, growth involves increase in cell mass and number of ribosomes, duplication of the bacterial chromosome, synthesis of new cell wall and plasma membrane, partitioning of the two chromosomes, septum formation, and bacterium division. This asexual process of reproduction is called binary fission. We call it here - reproduction or division.
2. The ability to growth depends on the "health factor" reflecting bacterium fitness, which we define as a linear function of the amount of nutrients consumed by individual.
3. For any bacterium to be propagated is necessary to provide the appropriate biochemical and biophysical environment. The biochemical (nutritional) environment is made available as a culture medium. This culture medium is represented by 2-D rectangular lattice of cellular automata with the Moor neighborhood, that is, the eight closest cells to the central one define its neighborhood.
4. We employ the "checker board" periodic boundary conditions [Dzwinel et. al., 1991] to model hexagonal box shape.
5. Motionless bacterium is located inside a single cell of the lattice.
6. Every organism must find in its environment all of the substances required for energy generation and cellular biosynthesis. The chemicals and elements of this environment that are utilized for bacterial growth are referred to as nutrients or nutritional requirements.
7. The cell is a source of infinite amount of nutrients and other growth factors (purines and pirimidines, amino acids and vitamins).

8. The cell with bacteria inside does not contain any nutrients.
9. In a single time step each bacterium consumes a fixed amount of nutrients from the neighboring cells.
10. Bacterial cells almost invariably take one of three forms: oblate (bacillus), sphere (coccus), or spiral (spirilla and spirochetes). We have assumed that the bacterium is oblate and motionless but it can change its orientation, which is quantized as shown in Fig.2:
    a. Horizontal (east – west)
    b. Vertical (north – south)
    c. North – east – south – west
    d. North – west – south – east

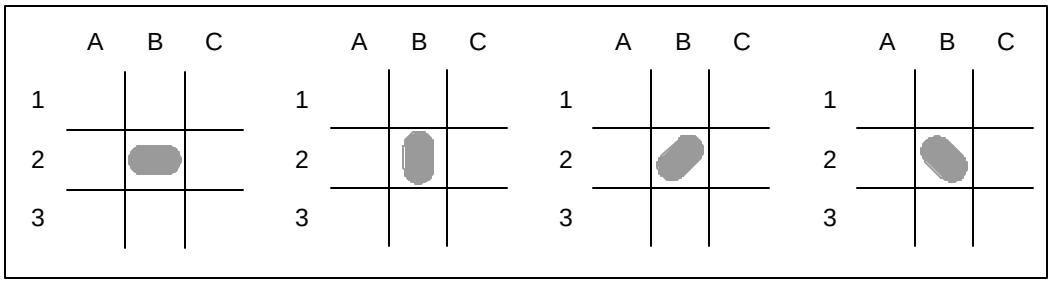

Figure 2 Possible orientations of bacterium

11. The ability of reproduction depends on the amount of free space in bacterium neighborhood.
12. The bacterium can divide producing a clone provided that its "health factor" is greater than a presumed value. The clone can occupy one of the neighboring cells. As displayed in Fig.3, its position is related to the parent orientation.

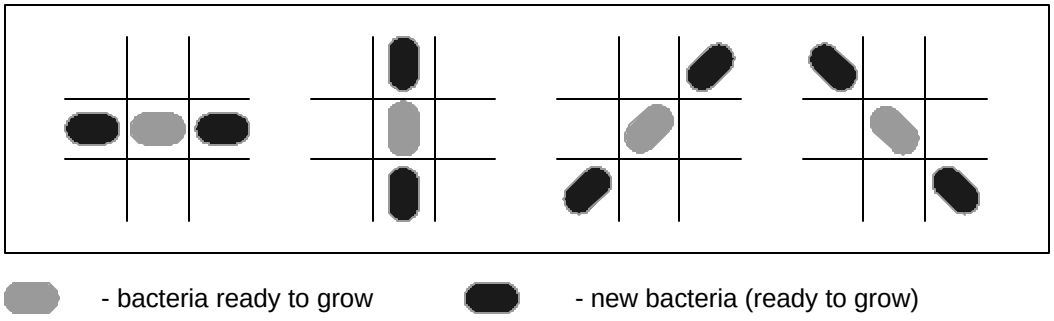

Figure 3 Bacteria orientation and its relation to its clone position

13. If bacterium "health factor" is low enough, it can die or morph into resting spore with a given probability.
14. The resting spore can morph back into living organism in favorable conditions. After dehibernation its orientation is chosen randomly.

The procedure executed by the agent, who represents a bacterium is displayed in Fig.4. In Table 1 and Table 2 we have collected the principal parameters describing bacterium and the environment, respectively.

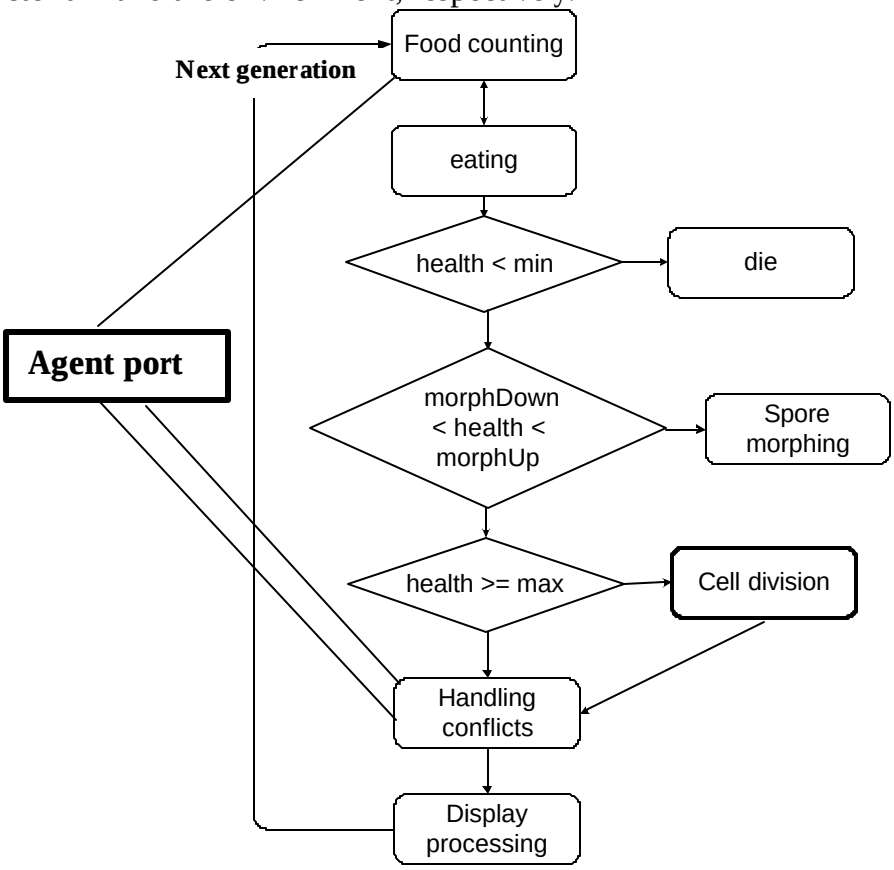

Figure 4 The procedure executed by the agent (bacterium)

Table 1 The principal parameters of the bacterium model

The name of parameter	Meaning
Orientation	There are four bacteria orientations shown in Fig.2
Health	The health factor of bacterium
IncrementHealth	An incremental value, which is added to the health factor of a bacterium, which consumes the maximum amount of food from its neighboring cells
SaveHealth	An incremental value, which is added to the health factor of a bacterium, which consumes **half** of the maximum amount of food from its neighboring cells
DecrementHealth	An decremented value, which is subtracted from the health factor of a bacterium, which is starving – no nutrients available in the neighborhood
WantsToGrowHealth	Determines if bacterium is ready to divide or not. If Health ≥ WantsToGrowHealth then bacteria is ready to produce a clone

WantsToMorphHealthDown, *WantsToMorphHealthUp*	Defines the range of health factor value for which bacterium can morph into a spore with a given probability.
WantsToMorphProbability	Probability of morphing
XPosition, *YPosition*	Bacterium position on 2D lattice, 720×416
MaxProbability, *MedProbability*, *MinProbability*	Probability of producing a clone in the most and less favorable directions according to the orientation of the parent bacterium

Table 2 The principal parameters of the environment

The name of parameter	Meaning
GridSizeX, *GridSizeY*	The size of the lattice
InitialNumberOfBacteria	Initial number of cells occupied by bacteria
InitSpores	Logical value. .*True.* means that the system allows for creating spores.

In Fig.5 we display the Moor neighborhood and "checker board" periodic boundary conditions (CBPBC). These boundary conditions described in [Dzwinel et. al. 1991] allows for modeling different shapes of computational box. Therefore, instead of a square box consisting of N cells, one can model by using CBPBC the optional box shape made of the same number of cells, including the most favorable - hexagonal box. The area of the sphere inscribed in hexagon is about 15% greater than in the square thus the bacterial colony can be simulated for a longer time by using the same memory load.

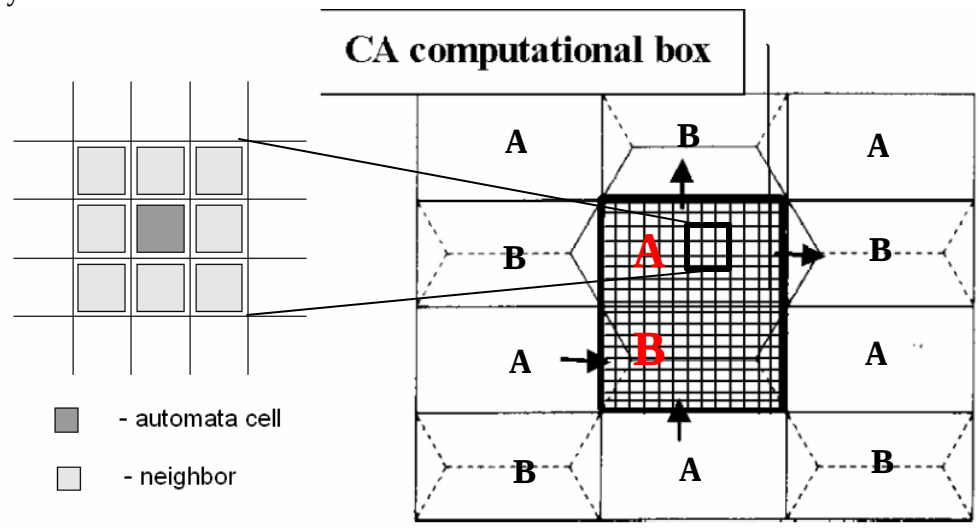

Figure 5 The Moor neighborhood at left and "checker board" periodic boundary conditions at right. The computational box consists of two sub-boxes A and B, which are periodic in Y direction and cross-periodic in X direction. Appropriate choice of the aspect ratio for the sides of the rectangular sub-boxes allows simulating hexagonal shape of the computational box.

We have assumed that each cell contains infinite amount of nutrients. The only empty cells are those which are occupied by bacteria and the cells surrounded by predefined amount of bacteria *n*. The value of $n_i \in \{2,...,8\}$ is set for every cell *i* with a given predefined probability. It means that if the number of bacteria is greater or equal to n_i, the cell is empty and does not contain nutrients. A few examples of empty cells are shown in Fig.6 for $n=2$ and n=4.

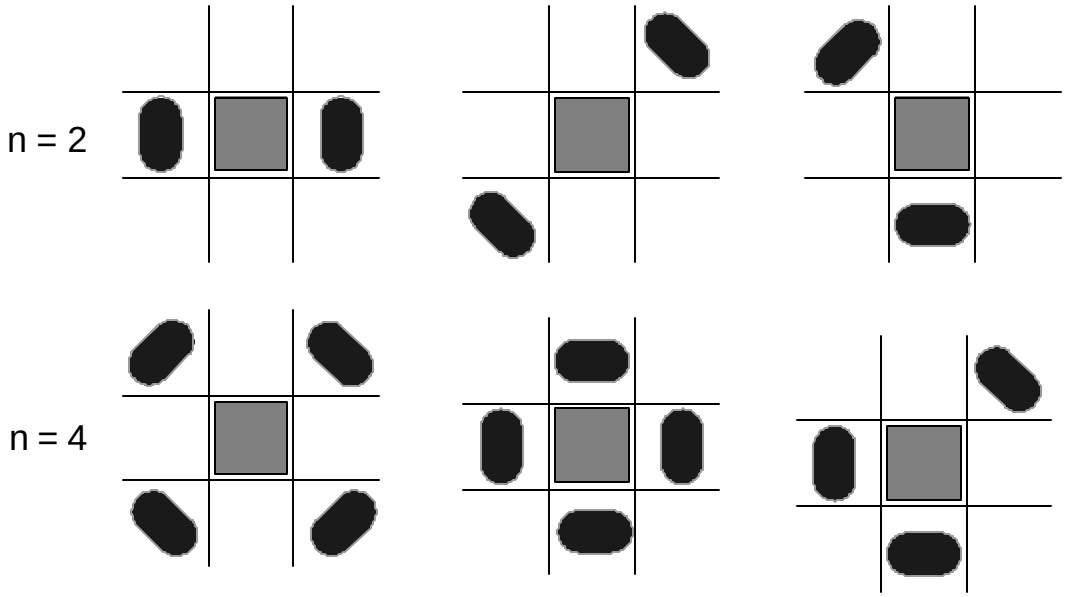

Figure 6 Empty cells in food applying algorithm

If the amount of nutrients consumed by bacterium is large enough it can divide producing one clone. Since only one individual can occupy one cell the location for the clone has to be found. We have assumed that the clone has to be situated in the closest neighborhood of its parent. The simplest scheme to handle bacterium division is as follows:

1. Chose one bacterium from those which are ready to divide
2. Select a cell in its neighborhood by using random number generator
3. Create a new individual and place it in the cell selected
4. Go to the step 1

The conflict will occur, if the selected location is already occupied. Therefore, the algorithm has to exclude such the cells from the selection. The following type of conflict arises when two parents choose the same cell for their clones. We can assume that in this case the strongest clone survives.

This scheme cannot produce the colonies for specific organisms shown in Fig.1 and consisting of chains or rod-like clusters, such as the anthrax. To solve this problem we define three additional values MIN, MAX and MED for each dividing bacterium. They represent the probabilities for locating the clone in the neighboring cells situated in one out of three different orientations according to the orientation of the clone parent. Since the anthrax bacterium divides along its shorter side the choice

of different cell than MAX/2 (see Fig.7) for a new clone, means that its bacteria parent must rotate earlier, as shown in Fig.8.

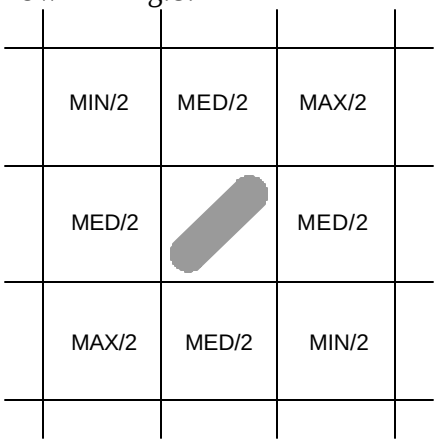

Figure 7 Growth probabilities assigned to locations

An improved algorithm capable of realizing bacterium replication can be decomposed as follows:

1. Choose all the bacteria, which are ready to divide.
2. For each dividing bacterium calculate the location of its clone according to the probabilities MIN, MED and MAX (see Fig.7).
3. If the selected location is occupied already the reproduction is congested.
4. If the location is empty and it is selected only by one clone, place the clone there.
5. If the location is selected by more than one clone the strongest clone wins.
6. If the clone is located in MIN/2 or MED/2 cells (see Fig.7), rotate its parent as shown in Fig.8.

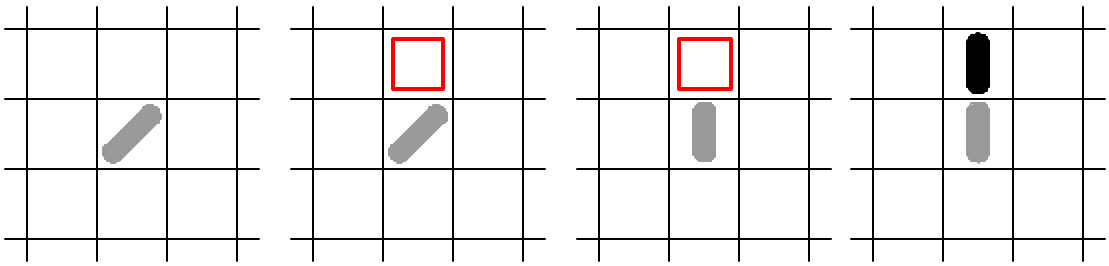

Figure 8 An example of dividing bacteria. a) Parent bacterium (gray) b) location calculated (red box), c) rotation of the parent d) new clone creation (clone is black)

4 Results from modeling

The computational box is represented by a rectangular consisting of 3×10^5 cells. The aspect ratio of the box sides is selected in such a way that the rectangular models the periodic hexagonal box by using “checker board” periodic boundary conditions

[Dzwinel et. al., 1991]. We have assumed that the incremental value *IncrementHealth* (see Table 1), which is added to the health factor of bacterium, which consumes optimal amount of nutrients, is two times greater than the penalty factor *DecrementHealth*, which is subtracted from the health factor of starving bacterium. This is natural for living organisms that a hostile environment influences them much stronger then the friendly conditions built them up. Moreover, such the assumption allows for faster response of the entire population on its local density fluctuations. We have assumed also realistic parameters for morphing. In Table 3 we collect all parameters taken for modeling, which are identical for each simulation.

Table 3 The values of parameters, which are common for all simulations

Parameter	Value
GridSizeX	720
GridSizeY	416
IncrementHealth	1
DecrementHealth	2
SaveHealth	0
WantsToMorphHealthDown	2
WantsToMorphHealthUp	3
WantToMorphRandom	0.3

The goal of modeling is to find the most favorable conditions for which average health factor for the population is the highest. We have simulated two groups of populations. The first group enables morphing (*initSpores*=.True.) and the second not (*initSpores*=.False.). We also investigate two types of bacteria features, which influence the population growth for the both groups:

1. the value of health factor for which bacterium is able to produce a clone (*wantsToGrowHealth*),
2. the values of MIN, MED and MAX probabilities (see Figs.7,8), which decide about the orientation of a new born clone in relation to the orientation of its parent.

In Table 4 we collect the parameters for all populations simulated. All the populations were initiated by a single bacterium placed in the center of the computational box.

In Fig.9 we demonstrate the snapshots from simulations of the populations preventing the formation of spores. The patterns obtained for the populations with spores are very similar. From Figs.10 and 11 we can figure out that the populations No.2, No.8 (*initSpores*=.False.) and No.1, No.7 (*initSpores*=.True.) (see Table 4) have the greatest average health factor reflecting bacterium fitness defined as a linear function of the amount of nutrients consumed by individual.

At the same time, the bacteria from these populations form the longest thread-like clusters (see Figs.9,12,13). Nevertheless, the health factor and the clusters are greater for populations which prevent morphing (No.2 and 8). The spores have very small health factor and occupy the space, which cannot be populated and nour-

ished. This causes local increase in population density, which inevitably decreases fitness factor for the entire bacterial colony. For the rest of modeled colonies (see Table 4) the differences in the average health factors and the clusters sizes are meaningless.

Table 4 The parameters taken for respective simulations

Simulation number	initSpores	MAXprobability	MEDprobability	MINprobability	wantsToGrowHealth
1	true	0,94	0,05	0,01	3
2	false	0,94	0,05	0,01	3
3	true	0,6	0,3	0,1	3
4	false	0,6	0,3	0,1	3
5	true	0,25	0,5	0,25	3
6	false	0,25	0,5	0,25	3
7	true	0,94	0,05	0,01	7
8	false	0,94	0,05	0,01	7
9	true	0,6	0,3	0,1	7
10	false	0,6	0,3	0,1	7
11	true	0,25	0,5	0,25	7
12	false	0,25	0,5	0,25	7

By comparing Figs.10 to Figs.11 we can notice that the average health factor for populations in which "stronger" individuals can produce clones (*wantsToGrowHealth=7*) is greater than for populations in which also weaker individuals can divide, i.e., *wantsToGrowHealth=3* (see Table 1). On the other hand, the average individual needs more time for reproduction for the first case, thus the population with *wantsToGrowHealth=7* grows slower. The average and maximum cluster sizes are very similar for the two cases. For stronger population the difference in health factors between spores and living organisms is greater. Therefore, the gap between average health factors for colonies disabling and enabling formation of spores is much wider for *wantsToGrowHealth=7* case than for *wantsToGrowHealth=3*. All of these findings concerns only the colonies forming long threads such as No.1,2,7,8. For the rest ones, these differences are insignificant.

The number of individuals for all the populations grows in time as t^{2+a} where a is close to 0. As shown in Fig.10a,11a, in the most cases, the number of individuals increases faster for weaker populations with smaller average health factor. But this is not the rule. For example, in bacteria colonies, which snapshots from simulations are

displayed in Fig.12,13, the number of individuals forming rod-like structures can over perform populations with a smaller health factor. This is clearly depicted in Fig.14a. The population density is much greater for weaker populations, e.g., the densities of colonies from Fig.12 and Fig.13 are 0.65 and 0.49, respectively (see Fig.14b). The density of the rod-like population is very variable at the beginning of simulation.

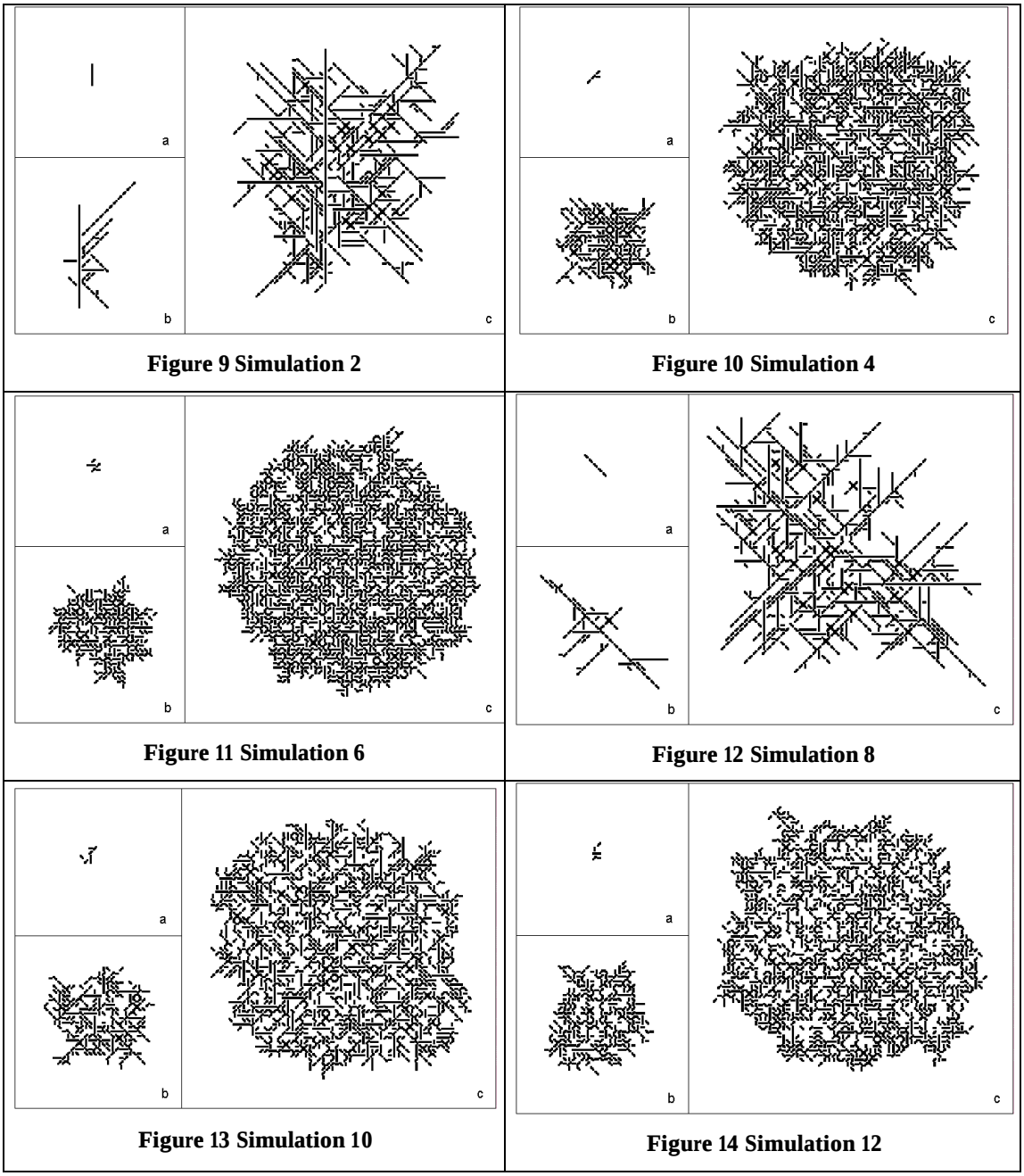

Figure 9 Simulation 2

Figure 10 Simulation 4

Figure 11 Simulation 6

Figure 12 Simulation 8

Figure 13 Simulation 10

Figure 14 Simulation 12

Figure 9 The snapshots representing a) 10, b) 50 and c) 100 generation from simulations of complex non-linear morphology of bacterial colonies. The parameters for respective simulations are collected in Table 4 (initSpores=.False.)

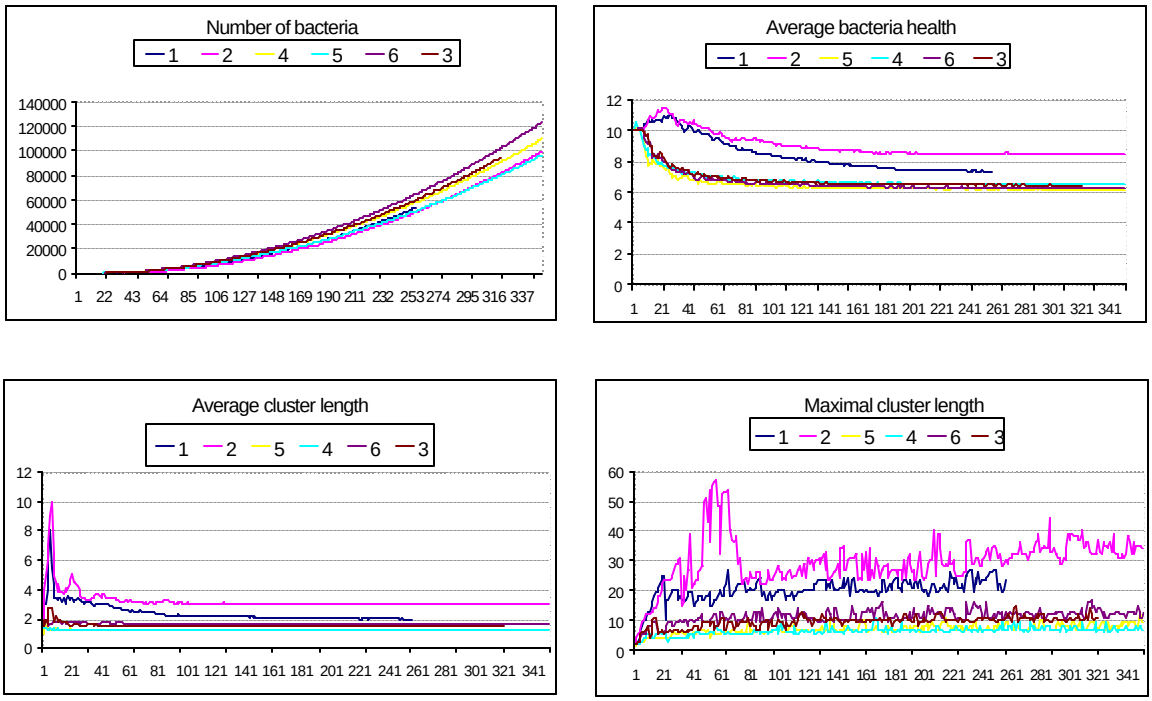

Figure 10 Results from simulation of the populations with a smaller value of the health factor for which bacterium is able to produce a clone (*wantsToGrowHealth=3*). The respective factors are shown in function of consecutive generations representing time.

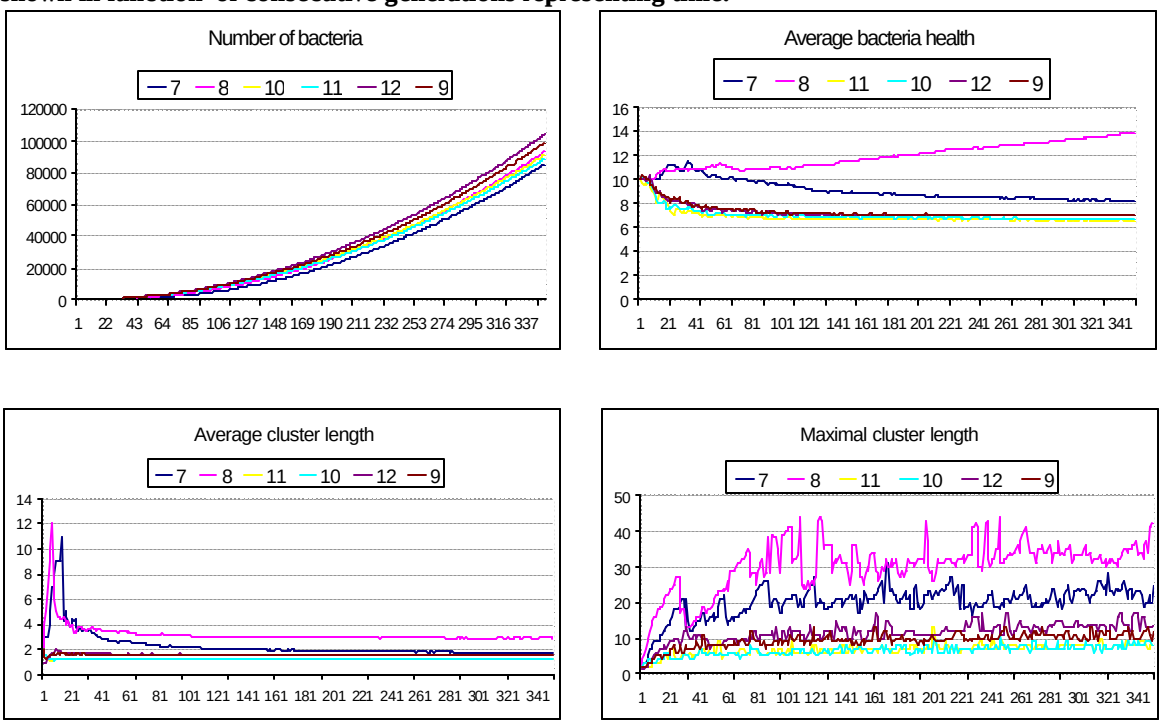

Figure 11 Results from simulation of the populations with a larger value of the health factor for which bacterium is able to produce a clone (*wantsToGrowHealth=7*). The respective factors are shown as a function of consecutive generations representing time.

It stabilizes after about 100 generations. This is due to difficulty in detection of the boundary of this colony. The rod-like colonies spread out much faster than the others. However, due to anisotropy of the rectangular grid the pattern produced by the population shown in Fig.13 is not circular one. The fractal dimension D computed from the formula $N(t)=R(t)^D$ where R(t) is the variable radius of the colony, for the two simulations from Fig.12 and Fig.13 is equal to 2.1 and 2.25, respectively. Because it is greater than 2 it means that the population still does not stabilize and the number of bacteria increases faster with *R(t)* than the number of cells encircled by its border.

Comparing the patterns presented in Fig.12 and Fig.13, one can discern that the population from Fig.12 is encapsulated in a shell-like structure. For more realistic model of environment, in which nutrients undergo diffusion, this skin can prevent nutrients to nourish the entire population. The structure of bacterial colony from Fig.13 is more permeable, allowing nutrients penetrate better into the interior of the population.

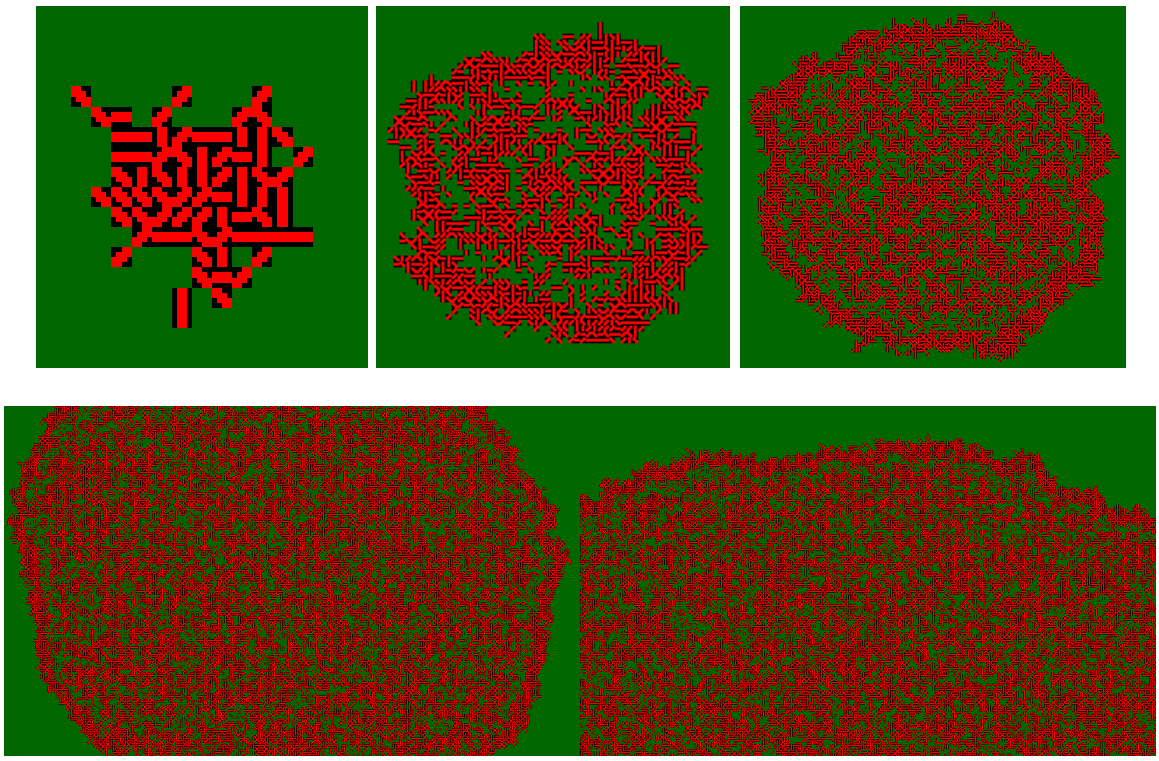

Figure 12 Snapshots from modeling of population No.5 (generations 10, 50, 100, 200, 400)

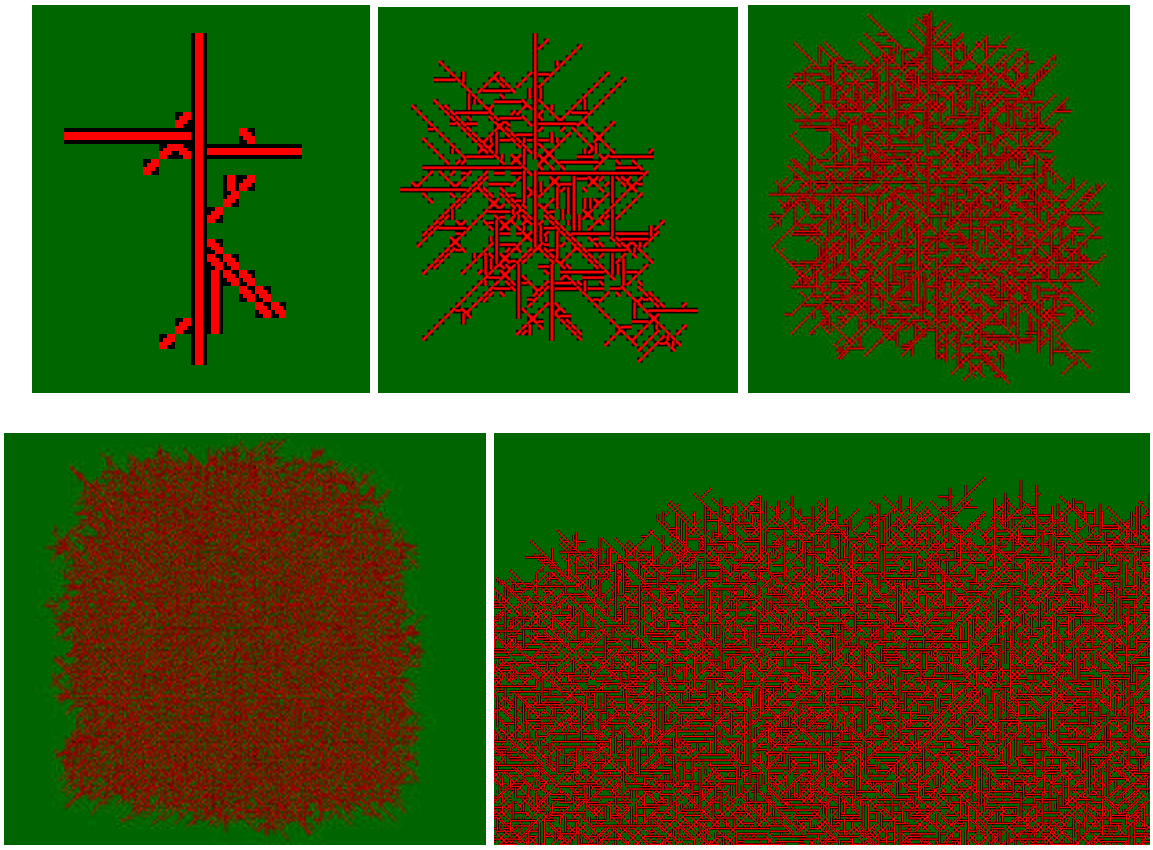

Figure 13 Snapshots from modeling of population No.1 (generations 20, 50, 100, 200, 400)

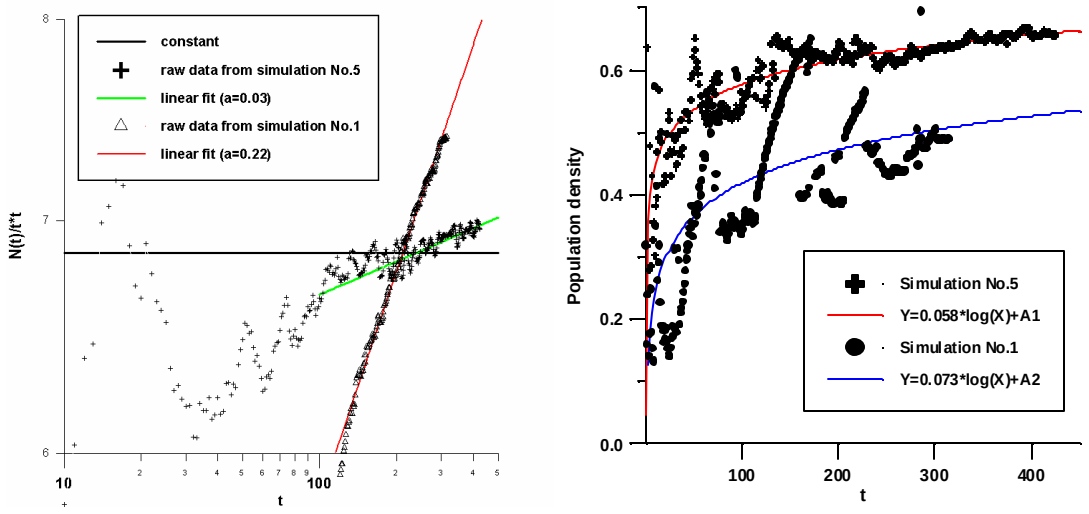

Figure 14 The plot displaying a) the power law of growth of the normalized population size N(t) and b) the density of colonies for simulations No.1 and No.5 from Table 4. The value of t is the time represented by the number of the generation.

5 Concluding remarks

The organisms living in large colonies such as: a human, ant, bee, coral and bacterium produce macroscopic patterns of various complexity. These non-linear patterns reflect different properties of the colonies including their adaptation features and fitness factors. The large-scale behavior comes from microscopic rules resulting from morphology of organisms and interactions of each individual with its neighborhood consisting of its physical environment and other individuals from the population. In our model the agents represent simple organisms such as bacteria, which principal goal is to grow and multiply. We have assumed that the bacterium cannot move but it can change its orientation on the rectangular lattice, which mimic an environment. The environment is made of identical cellular automata containing nutrients. Because the amount of nutrients in a cell depends on the presence or absence of bacteria inside the cell or in its closest neighborhood, the agents modify the environment. In turn, due to the feedback, the environment influences bacteria morphology (e.g., its reproduction ability, its health, death and morphing transformations), which is controlled by the availability of nutrients and the free space in bacterium neighborhood.

The optimal shape of bacterial colony increases population adaptability, which can be expressed by the value of its fitness factor computed as an average of "health factors" being linear function of nutrients consumed by a single bacterium. In our model this factor reflects the possibility to supply each of individual with as much nutrients, as is possible. The creation of the bamboo-like structures was forced by the microscopic rules of bacteria reproduction demonstrated in Figs.7,8, which mean that the population of bacteria, which do not change too often their orientation will produce complex elongated and rod-like structures.

The results of simulations show that bacteria colonies, which produce rod-like structures, have the greatest fitness factors. They also spread-out on the lattice much faster than other populations. Therefore, we presume that these forms of bacteria colonies can represent the most dangerous infections such as the anthrax (see Fig.1). We have shown also that populations producing both the rod-like structures and spores have smaller average health factor than those in which morphing is forbidden. However, their health factor is still much higher than for other types of populations, which do not produce any rod-like clusters. Despite decreasing the health factor, the morphing gives a chance for bacterium to survive in hostile conditions.

We show also that the average health factor can be greater for the rod-like populations involving greater value of the threshold health factor, which is required to initiate the reproduction process. However, in this case, the population grows considerably slower. The morphing slows it down additionally decreasing considerably the health factor for the entire colony. This is interesting that both morphing and reproduction threshold do not influence considerably the populations, which do not produce rod-like clusters.

Because our model is oversimplified, the patterns we simulate differ from those observed in laboratories. This is mainly due to regular structure of the lattice. To make the patterns more realistic we plan to use irregular mesh and simulate diffu-

sion of nutrients on this mesh. Also the health factor computed as an increasing function of nutrients consumed is not a good indicator of the population adaptability. It does not include information about ability of morphing as an important factor of populations' survival and adaptability.

The understanding of complex collective behavior of simple organisms is a key factor of development new technologies in nano-biology and nano-mechanics. The functions of agent object can be extended by introducing additional individual features such as leadership, intuition and creativity, which enables to employ this methodology also in the social sciences and economy [DeAngelis and Gross, 1992; Jager, 2000].

Acknowledgements

Support for this work has come from the Polish Committee for Scientific Research (KBN) project 4T11F02022 and AGH Institute of Computer Science internal funds.

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