Dynamical strategies for obstacle avoidance during *Dictyostelium discoideum* aggregation: a Multi-agent system model.

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Abstract

Chemotaxis, the movement of an organism in response to a chemical stimulus, is a typical feature of many microbiological systems. In particular, the social amoeba *Disctyostelium discoideum* is widely used as a model organism, but it is not still clear how it behaves in heterogeneous environments. A few models focused on mechanical features have already addressed the question; however, we propose that phenomenological models focusing on the dynamics may provide new meaningful data. Consequently, by means of a specific Multi-agent system model, we studied the dynamical features emerging from complex social interactions among individuals belonging to amoeboids colonies.

After defining an appropriate metric to perform meaningful measurements, we found that: a) obstacles play the role of local topological perturbation, as they alter the flux of chemical signals; b) that obstacle that physically block the cellular motion as well as the chemicals elicit dynamical evolutions that are not statistically distinguishable from the case where obstacles that do not interfere physically with said motion; c) that fluctuations of the dynamics can lead to better exploration of the physical space, thus preventing multiple stable aggregates.

From previous results, we may speculate about the fact that chemotactic cells, in many cases, can avoid obstacles by simply following the altered chemical gradient: social interactions seem to be sufficient to guarantee the aggregation of the whole colony past numerous obstacles. It is then unlikely that cells have developed special mechanisms to cope with the presence of topological perturbation sources. Nevertheless, we suggest that further studies can provide better understanding and that, in order to gain deeper knowledge, mechanical models should be coupled with phenomenological, system-oriented ones.

1 Background

The social amoeba *Dictyostelium dicoideum*, a model organism in biomedical research [1, 2], is a well-studied example of chemotactic life-form [3–8]. Chemotaxis involves the detection of local gradients of chemical signals (spatial detection), the polarization of the cell and the subsequent movement of the cell up the gradient [9, 10]. In particular, starving cells of *D. discoideum* are able to send and to process periodic stimuli of 3',5'-cyclic adenosine monophosphate (cAMP) that acts as chemoattractor [11–14]. As a consequence, a scattered colony is able to self-coordinate towards an aggregate bulk [15, 16].

Since directed migration is a common feature in many cell systems, *D. discoideum* is widely studied [17] and has inspired problems of decentralized gathering [18, 19]. Consequently, over the years, many aspects of chemotactic behavior within amoebozoan colonies have been studied both from an experimental [14, 20–22] and a theoretical [23–30] point of view. However, most of them only focus on homogeneous environments, while, on the contrary, heterogeneous environments are typical of *in vivo* conditions. A few studies have already tackled such issue [31, 32]; however, what was analyzed with individual-based models were the mechanical and rheological properties of the microbiological system. As far as we know, the only attempt to simulate the behavior of a whole colony in a heterogeneous environment with discrete models at an appropriate scale makes use of the Cellular Automata method [33, 34]. However, sometimes such work lacks the needed complexity level. By using a Multi-agent system [35, 36] model, the purpose of this article is to study the efficiency of chemotaxias in achieving controlled cell migration from a complex systems perspective [37, 38], thus focusing on dynamical

features and self-organization [39].

This paper is structured as follows: first, we defend the model choice and highlight the point-of-view shift occurring in phenomenological simulations like ours. Second, we define a quantitative variable that is appropriate to measure a complex colony evolution without direct dependence on the environmental topology. Third, we present the results: we study the obstacle-avoidance efficiency as a function of the aggregation efficacy of the colony as a whole and of the type of obstacle. Obstacles can either be "physical" (they block both cells and signals, for which they act as a sink) or "chemical" (they let cells through, but block signals). Finally, we speculate about the effect of noise on the probability of metastable clusters to appear by providing preliminary results. The to-be-presented approach can be effective both in a single-obstacle and a multi-obstacle environment, thus helping researchers to explore the entire space of configurations.

2 Methodology

Analytically speaking, obstacles are a source of topological perturbation: they alter the Euclidean plane according to their size, shape and position. Consequently, they introduce local border conditions. In particular, "physical" obstacles are represented by reflecting (local) boundary conditions, while "chemical" obstacles do not alter the physical plane. In plain English, it means that "physical" obstacles prevent amoebas from crossing them, while "chemical" ones do not, albeit interfering with the signals. Moreover, environmental heterogeneity occurs on a scale comparable to that of individual cells [32]. As a consequence, macroscopic continuum models (ODEs) of cell movement are not appropriate or have great difficulties in tackling the problem of heterogeneous domains [40].

On the other hand, individual-based models that focus on single cells neglect the complexity of the system: they effectively inquire what happens to the cell movement in response to chemical gradients $\nabla C = g\,\hat{z}$ (after having appropriately oriented the coordinates) around an obstacle, but they do not consider the dynamical interactions among multiple cells behaving as a complex network [41–43]. In fact, *D. discoideum* colonies consist of a population of simple agents interacting locally with one another and with their environment. Although there is no centralized control structure dictating how individual agents should behave, local interactions (even random ones, up to a certain degree) between such agents may lead to the emergence of global complex behavior [44]. Consequently, another modeling approach may better analyze those dynamical features, such as positive feedback loops or multi-stability [45], that typically let behaviors such as collective choices and self-organization to emerge [46]. In the present context, we may conjecture that mutual interactions among cells can lead to (or, at least, contribute to) coordinated cell movement and obstacle avoidance.

Consequently, we may need a meso-scale approach, which involves discrete and individual-based modeling and that focuses on the colony as a whole and not on a limited number of cells. Moreover, it should be constructed so that we can analyze the dynamical properties of the system. What we believe to be the best approach that adheres to the listed requirements is the Multi Agent System paradigm [35, 47].

2.1 The chosen MAS model

In order to perform simulations and analysis, we make use of an existing MAS-based computational framework that was purposefully designed to address *D. discoideum* behavior and that has already been tested and validated [48]. Recall that such model involves the generation of the desired dynamic after stating individual behavioral rules and parameters. Four main agents types compose the model architecture: Environment Env (a squared closed $\dim \times \dim$ domain, composed by cells with associated food sources $b(t)$ possibly growing over time), Amoeba Am (proactive agents representing individual cells), cAMP (vectorial packages of chemical signal) and Obstacles Obs. It has been shown [48] that the vectorial message-sharing approach generates results that are consistent to those obtained with the typical diffusion of chemicals, as we can substitute a chemical gradient $\nabla C = g\,\hat{z}$ with one that is associated to the flux of probability of sensing a discrete signal: $P^{amoeba}(\{I_{cAMP}^{starving} : E_g \mapsto \mathrm{per}\})$, that is, the probability of a (starving) moving amoeba processing the information carried by an absorbed cAMP message. At the same time, such strategy is cheaper in terms of computational cost than diffusive ones [49, 50]. In addition, stochastic perturbations and individual failures can be added easily and controlled by tuning

the respective parameters.

The model was carefully examined and validated by identifying its scale-invariant region (with respect to its microscopic variable N, number of amoebas, and the macroscopic one ρ, density of amoebas), by quantifying its associated systematic errors and by comparing its results to those obtained by other models and the biological literature. Although it only focuses on the first aggregating stages of the gathering process, and it does not implement adhesion forces between neighbor cells, we believe that said model could represent an appropriate tool to investigate the dynamical properties of a complex colony while coping with topological perturbations due to obstacles.
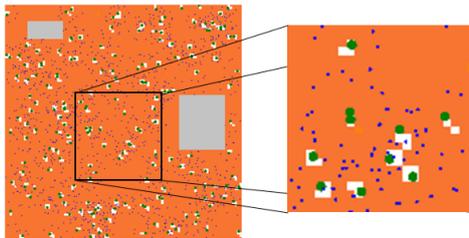
Figure 1: Representation of a typical setting: a squared environment with constant food sources (orange) contains agent Amoeba (green) that is able to reproduce the sending-sensing-orienting-moving behavior that characterize chemotactical motion. Blue dots are cAMP agents, while gray objects are obstacles.

2.2 Defining quantitative measurements

In the present section, we suggest a "mean local gathering factor" metric in order to quantify the aggregation stages of the colony. In particular, it should have the following properties: it has to be a monotonically increasing function of time t if the colony is steadily aggregating and it must not be directly dependent on the environmental topology. Thanks to those properties, it is suitable a) to quantitatively assess the evolution of the dynamics over time (as a function of the aggregation rate of the whole colony) and b) to cope with a heterogeneous environment. The metric is defined as follows:

1. We define a certain range (in turn defining a "neighborhood area") under which two cells can be said to be neighbors. The reason for defining a "range" is that, as adhesion cells are not implemented in the model, cells are kept close by social interactions (message sharing) [48]. As a consequence, two neighbor cells can vary their distance in time by oscillating around each other. For each agent, said "neighborhood range" $R_N \in \mathbb{R}$ is defined as follows:

$$R_N = R + v_A \cdot \Delta t - r$$

   where R is the agent radius, v_A is the agent speed, Δt is equal to 1 cycle; r is the mean distance from the centers of two overlapping cells, that can be determined by studying the chosen simulations. Agent parameters and exact values come from settings described in [48].

2. In the position space, the "neighborhood area" of the i-th cell is given by:

$$A_i^* = \{(x', y') \text{ s.t. } (x_i - x')^2 + (y_i - y')^2 \leq R_N^2\}. \tag{1}$$

   where (x_i, y_i) are the cell center coordinates. At the same time, the physical space occupied by the j-th agent is given by:

$$A_j = \{(x'', y'') \text{ s.t. } (x_j - x'')^2 + (y_j - y')^2 \leq R^2\}. \tag{2}$$

3. Given the i-th agent, the set $in(t)_i$ of j-amoebas that can be considered i-th neighbors (at time t) is the set of j-agents whose physical area intersects the i-th "neighbor area" at time t, that is:

$$in(t)_i = \{\{\text{Am}_j\}, j = 1 \ldots n \text{ s.t. } [\text{Am}_j \cap \text{Am}_i^*]_t \neq \emptyset\} \tag{3}$$

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4. The number of neighbor cell to the i-th amoeba is thus given by:

$$n_{in}(t)_i = \dim(in(t)_i) \tag{4}$$

and it represents the *local gathering factor*, as it increases the deeper the i-th amoeba is in the gathering patter and/or in the final aggregate.

5. To obtain a *mean local gathering factor*, we average $n_{in}(t)_i$ over the number of amoebas N:

$$\aleph(t) = \frac{1}{N} \sum_{i=1}^{N} n_{in}(t)_i \tag{5}$$

Note that $\aleph$ will not be normalized, since the maximum value it can reach at the end of the simulation time t_{max} is itself something informative about the "strength" of the gathering process. $\aleph$ is similar to an entropy-based function [51] and should be a

2.3 Simulation protocol and remarks

The analysis is based on the following protocol.

1. Formulate a hypothesis about what phenomenon is going to be inquired and what to expect. In the case of the present project, it will be focusing on the effect of obstacles placed into the environment: are their effect local or do they influence the whole system? How does their nature change the dynamics?

2. Set the simulations in a meaningful position in the parameter space $\mathbb{SP}$. The main aim is not to completely investigate such space, defined by the number, shape, location and nature of obstacles ($\mathbb{SP} = \{n_{obs} \times \hat{\dim} \times \bar{x}_0 \times \mathrm{nat}_{obs}\}$), but to verify whether a certain configuration elicits local adaptations of the dynamics. Therefore, such configuration is randomly chosen among those that respect the following requirement: that the location of the obstacles would be in a limited subspace $\mathrm{Sub}_{\mathrm{Env}} \subset \mathrm{Env}$ of the domain. This way we can compare what happens when obstacles are placed locally and whether the obstacle-free subspace elicits the same aggregation pattern as when $n_{obs} = 0$. Consequently, after having set the colony parameters according to those that were validated in paper [48], a point in $\mathbb{SP}' = \{n_{obs} \times \hat{\dim} \times \bar{x}_0' \in \mathrm{Sub}_{\mathrm{Env}} \times \mathrm{nat}_{obs}\}$ is chosen.

3. Set external noise sources to zero, since the main focus is on perturbation caused by obstacles.

4. Run repeated simulations of early aggregation in order to increase statistic relevance while measuring the defined variable $\aleph(t)$. Meaningful parameters and variables are set according to those validated in paper [48] (see Table 1). As the simulations described in [48] are specific for the pre-aggregation stages of the gathering process, we remain consistent in considering the same time interval for the analysis.

Table 1: Meaningful parameters and variables for simulations (set to be biologically consistent) [48].

$\rho[\frac{\mathrm{cell}}{mm^2}]$	N	$v_A[\frac{\mathrm{unit}}{s}]$	$v_c[\frac{\mathrm{unit}}{s}]$	t_S [s]	P_A
563	2640	1.4	4.7	10	0.001

5. Study the influence of nat_{obs}.

3 Obstacles as local source of perturbation

By following the aforementioned protocol, we first inquired whether obstacles represent local sources of perturbation. We run repeated simulations of the same setting, which was randomly chosen among $\mathbb{SP}$, thus measuring ($\aleph \pm \sigma_\aleph$) and, at the same time, looking at simulated animations. nat_{obs} was varied

from "physical" to "chemical".

In fact, obstacles perturb the colony dynamics, but only locally: as it can be seen in Fig. 2, they circumscribe different subspaces with different concentrations of cells, thus eliciting metastable clusters, but, where they are absent, it can be seen the usual (namely, the same as registered in other modeling papers [24, 26, 48]) behavior towards the aggregate. Moreover, as we can notice in Fig. 3, the measure $\aleph$ is indeed monotonically increasing, as we would expect in case of a steadily aggregating colony.
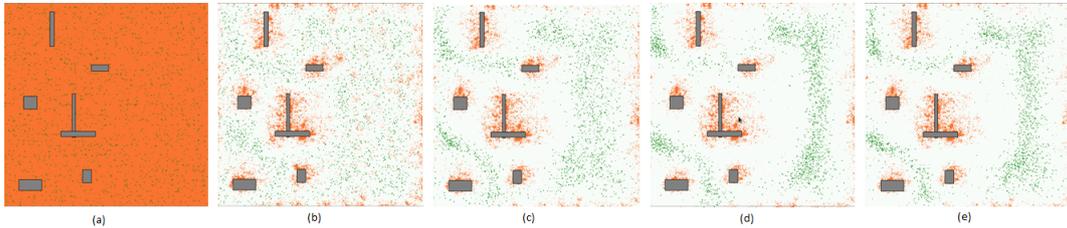
Figure 2: Evolution of the colony from animation. It refers to the same experiment whose measurement is in Fig. 3. (a) setting; (b) t =400, right after the (conventional) transient threshold; (c) t = 700, we can see different areas with different cell density; (d) t = 1000, multiple density peaks are quite clear within Sub_{Env}, while the half environment without obstacles, although its shape is different (rectangular instead of squared) from the "usual" one, the colony here is streaming and gathering; (e) t = 1300 the metastable clusters are even more recognizable. Note that, during the evolution, all bacteria have been eaten.
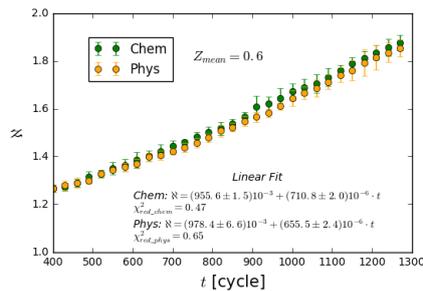
Figure 3: Evolution during early aggregation with chemical and physical obstacles. The time interval is consistent with that validated in [48]. ($\aleph \pm \sigma_{aleph}$) is indeed monotonically increasing in time, as we would expect in case of a steadily aggregating colony (curve fit are not shown for clarity reasons). Many different simulations with different obstacle configurations $\in \mathbb{SP}$ gave the very same qualitative result. Moreover, a Z test confirms that the two experiments are not statistically separate.

4 Emerging choices for obstacle avoidance

From a computational point of view, interpreting an obstacle as "physical" or "chemical" is equal to saying that a single cell has or has not, respectively, a special obstacle-sensing mechanism. In fact, physical obstacles are such if cells are able to sense their presence and to steer away from them. On the contrary, in settings with chemical obstacles that perturb the chemical flux, cells are not required to own particular sensing mechanisms, as they can in many cases avoid the obstacle by simply following the perturbed chemical gradient in its vicinity. Such hypothesis was already tested with round obstacles and single-individual-based models [31, 32]. However, we suggest that similar results can be obtained when considering the whole complex system: nonlinear interactions among cells lead to collective choices, such as obstacle avoidance from one side or the other, through bifurcations that qualitatively change the dynamics and lead to new attractors. The simplest way to model said phenomenon would therefore be by using positive feedback loops: the more cells choose a certain avoiding path, the more the probability

flux of signals increase, thus the more other cells will choose the very same path.

The very same phenomenon can be observed in MAS simulations, where the complexity of the system is better taken into account and where multiple obstacles with relaxed conditions can be placed and tested. Fig. 3 already showed how "physical" and "chemical" obstacles perturb the dynamics in a way that is statistically indistinguishable. Other settings in the parameter space $\mathbb{SP}$ give the very same qualitative result. Then, it seems that it is not necessary for a cell to develop specific mechanism for obstacle avoidance: it is guided by chemical messages that, once being been absorbed by obstacles, highlight the best paths for chemotactical migration through social interactions. It means that the self-aggregating patterns of the colony as a whole not only guide single cells towards stable clusters (attractors), but they also elicit bifurcation and collective choices in order to avoid obstacles.
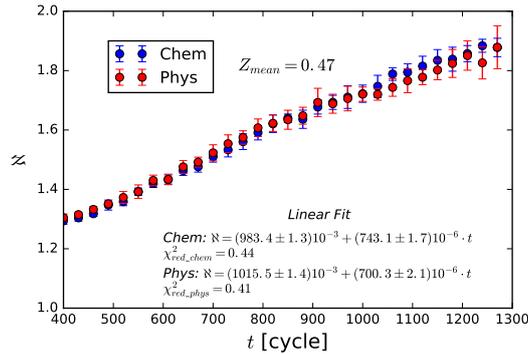
Figure 4: Evolution of $(\aleph \pm \sigma_\aleph)$ for both chemical and physical obstacles. Although not shown (for clarity reasons) a linear fit was performed guaranteeing the increasing trend; χ^2_{red} values are shown. A test Z, evaluating the mean distance between the two data set, was also performed and its mean value shown in the chart.
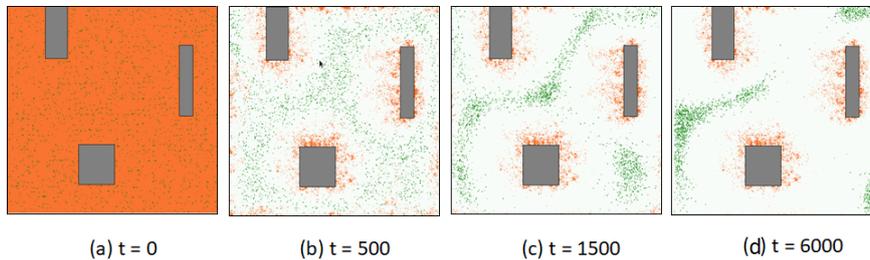
Figure 5: Several snapshots during the evolution of a colony. In order to magnify the multistable clusters, simulations were let run even after the aggregation interval. Obstacles were inserted into the domain. Snapshots refer to the experimental set whose measurements are shown in Fig. 3. (a) Experimental setting; (b) After an initial transient, streams appear; (c) Early aggregation is almost completed. Amoebas go on migrating; (d) Multistability is clear: there are three main attractors around which amoebas are gathering. Note that cells have explored almost all the physical space (they ate all bacteria that were deployed initially.)

4.1 Fluctuations in the dynamics

Fluctuations around a mean behavior are known to elicit better explorations of the state space [45,52]. Moreover, chemotactically-driven dynamics tend to make cells tighter with the initial cluster, while noise-drive dynamics allow cells to move in areas with prior lower cell density and, possibly, to guide streams and migrations by means of positive feedback loops. In fact, if setting other simulations on off-scale (noisy) configurations (for instance, $\rho = \frac{200}{100^2}$, but $N = 200$ [48]), it is less likely to observe the formation of multiple stable clusters (see Table 2).

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Table 2: Chances [%] to get multiple stable clusters (at least 2). Data from multiple (100) simulations with different obstacles configurations.

	on-scale	off-scale
1 obstacle	15	3
multiple obs	70	30

Instead of being stable, little clusters are metastable: they last as long as fluctuations do not drive single cells toward the cAMP flux that comes from the biggest cluster. After that, slowly but steadily, all the small clusters stream towards the biggest one after being guided by interactions with other migrating cells. Collective behavior towards bifurcation, then, seems to result from a delicate trade-off between robust chemotaxis and random fluctuations as suggested by previous experimental works [53].

5 Conclusions

The main aim of this project was to investigate how colonies of chemotactical cells behave when obstacles are present in their environment, as often happens *in vivo*. To do so, we chose a model organism, the social amoeba *Dictyostelium discoideum* and we studied its evolution during the aggregation process. Since we decided to focus on dynamical features elicited by nonlinear social interactions rather than on individual mechanical properties, we exploited a phenomenological Multi Agent System model that had been purposefully designed, implemented and validated. A suitable metric (the *local clustering factor* $\aleph$) was also suggested in order to measure the aggregation progresses without being directly dependent on the environmental topology.

The presence of obstacles perturbs the chemical flux. A group of cells in collision with an obstacle can in many cases avoid it by simply following signals coming from other directions. In fact, as far as the present model is concerned, whether agents Amoeba possess specific behaviors to avoid obstacles or not, they manage to self-aggregate without differences in the dynamics that are statistically relevant. A similar result suggests that, from a population point of view, social interactions are more effective than individual capabilities to detect and avoid obstacles. In fact, large and complex colonies are only locally perturbed by the presence of topological perturbation and seem to be able to cope with them thanks to the same behaviors that lead to aggregation in homogeneous environments: it was experimentally shown that the "usual" social interactions elicit collective choices, in order to avoid obstacles and stream towards the main aggregate.

Moreover, preliminary results suggest that a system whose dynamic is noisy can better explore the state space and overcome obstacles without (or with less probability) forming multiple stable clusters. On the other hand, of course, perturbed colonies are less likely to follow the chemical gradient, so an efficient gathering process should present a trade-off between chemotactical stability and random exploration as already suggested in the field of system control [54].

Often, organisms that live in swarms have little concern for an individual fate, whereas they are evolutionary competitive as a whole population [18]. Therefore, focusing on the entire colony instead of single individuals can provide additional information in order to understand specific behaviors. We believe that individual-based, although holistic MAS models can be useful when addressing complex microbiological systems in heterogeneous environments. In the present context, we were able to get interesting preliminary results from our phenomenological simulations that can be coupled with those from mechanical models; furthermore, we would suggest further studies in order to better explore the configuration space $\mathbb{SP}$ and to inquire the dependencies of the dynamics on the ratio between obstacle and cell (or population) size.

To summarize, it was shown that, as far as colonies of *D. discoideum* are concerned, emergent dynamical features are sufficient, from the whole colony point of view, to avoid obstacles placed in the domain. Additional individual abilities might elicit better efficiency, but such is not the case observed in the present simulations. This may explain why specialized biological mechanisms for avoiding obstacles are only known for a few cells and organisms [32]. Further *in vivo* and *in vitro* studies are recommended

to improve our knowledge on behavior of cellular motility in heterogeneous environments.

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