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# Macroscopic growth of aerobic bacteria

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#### **Abstract**

In this experimental and theoretical work, we introduce and explain the macroscopic growth of bacteria using an experimental, mathematical, and physical model combined with traditional microbiology.

The main objective of this original model is to provide experimental and theoretical evidence that the exponential spatio-temporal growth of a bacterial colony on a two-dimensional surface precisely follows the solution of the heat diffusion equation, including its source/sink term and boundary conditions.

In other words, we propose that the macroscopic growth of bacteria obeys physical and mathematical principles, particularly those governing heat diffusion and electromagnetism.

These electromagnetic fields present in bacterial colonies help to explain the complex patterns that form during macroscopic bacterial growth.

This shows that, in particular, magnetostatic bacteria, which contain iron-rich compounds, can detach themselves from the rest of the colony and follow magnetic field lines in concentric circles.

Clearly, these findings challenge the classical model of bacterial "walking" based on flagellar movement.

Furthermore, this work suggests that bacterial growth patterns can be modeled by partial differential equations, similar to those used to describe heat diffusion and electromagnetic fields within a closed control volume, as experimentally demonstrated by the formation of these patterns.

Indeed, experience proves that the growth and movement of a bacterial colony are influenced by its own intrinsic electric and magnetic fields, in addition to the sensation of heat.

It is clear that this description differs considerably from the traditional microbiological approach.

Traditional microbiology explains microscopic bacterial growth as the result of the multiplication of individual cells and their interactions with their environment at the microscopic scale. Our work, on the other hand, is precise and focuses on the macroscopic scale, suggesting that the collective behavior of millions of bacteria within a colony can be described using principles of mathematical physics combined with microbiology.

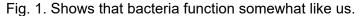
In summary, our research offers a mathematical physics perspective on bacterial growth, demonstrating that the spatio-temporal growth and organization of a bacterial colony can be accurately described by the same equations and principles that model physical phenomena such as diffusion and electromagnetism.

### **I.Introduction**

There are two main approaches to studying bacterial colony growth:

i- The traditional microbiological approach [1,3,4,5,6].

Indeed, the traditional approach focuses on cellular mechanisms, such as flagellar movement and cell replication, which govern temporal growth. This description is illustrated in Figures 1, 2, and 3 [1,4,5,6] below:



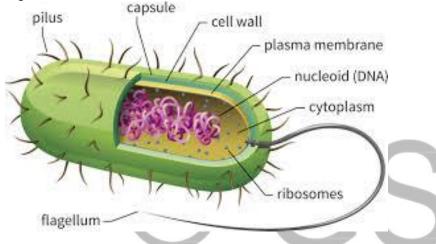
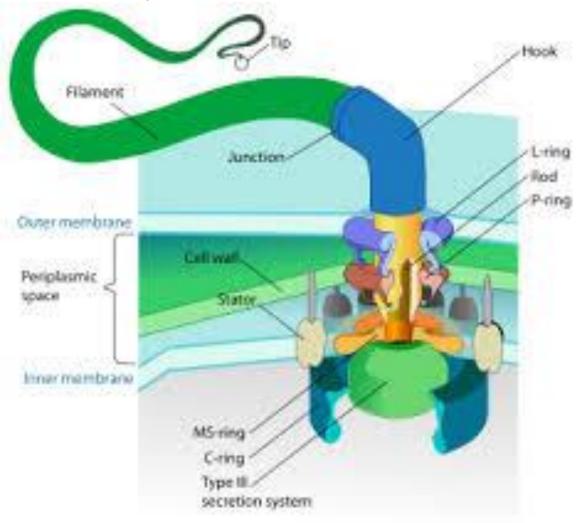


Fig. 1. Shows that bacteria function somewhat like us [1].

Fig. 2. Shows that bacteria have legs or flagella of some kind used for walking [1].



Colonies of bacterial cells are the main source of disease and food.

Bacterial microbes are unicellular microorganisms lacking a nuclear

membrane, metabolically active and when they grow, beyond a certain limit, under appropriate conditions of food, humidity and oxygen, they divide by binary fission. This means that the

microscopic growth of individual bacterial cells is ultimately arrested by the bacteria self-dividing into two daughter cells.

This process is called binary fission.

Provided no external effects occur, the resulting daughter cells are genetically identical to the parent cell. Obviously, this results in an exponential growth of bacterial colonies.

The temporal evolution of bacterial colonies goes through three main phases, namely the lag phase, the logarithmic or exponential growth phase and finally the exponential death or decay phase [1,2].

The physical interactions of developing bacterial cells with each other rand with their growth environment (Agar food, temperature, humidity, etc.), significantly affect the structure and movement dynamics of bacterial colony biofilms.

The currently burgeoning field of theoretical and experimental studies on bacteria, particularly on the movement or walking of bacterial cells, focuses on the role of their legs called falagellae Fig1.

Figure 1 shows the walking mechanism of bacterial cells, the role of the legs or falagellates in their transfer or walking has been the subject of previous work.

Katsumi Imada et al 2018 [3] states that many motile bacteria swim and swarm toward favorable environments using the flagellum, which is rotated by a motor embedded in the inner membrane.

The bacterial flagellum is a mobile organelle composed of thousands of protein subunits. He adds that Gram-negative bacteria, such as Escherichia coli and Salmonella, swim by rotating helical filamentous organelles called flagella. The flagellum rotates at a relatively high speed and is driven by a reversible rotary motor embedded in the cell membrane at the base of the filament.

Sandra Postel et al 2016 [4] stated that Flagella are crucial for bacterial motility and pathogenesis. The flagellar capping protein (FliD) regulates filament assembly by chaperoning and sorting flagellin (FliC) proteins after they traverse the hollow filament and exit the growing flagellumtip.

In the absence of FliD, flagella do not form, resulting in impaired motility and infectivity.

He asserts that "many bacteria, including several that cause disease in humans, have long, whip-like appendages called flagella that extend well beyond their cell walls.

Flagella can spin and propel bacteria through liquids, such as water or blood, and they are mostly made up of thousands of copies of a single protein called flagellin. During the construction of flagella, the flagellin proteins are placed in their appropriate positions by another protein called FliD, several copies of which form a cap at the end of the flagella. Without FliD, bacteria cannot properly assemble flagella and therefore cannot swim; it also hinders their ability to cause disease.

W. Wilson Nedeljkovic, 2021 [5] stated that the bacterial flagellum is one of the most complex and dynamic biological nanomachines known and has attracted attention since its discovery in the late 19th century. The history of flagellar research has gradually evolved from purely morphological studies to biochemical and biophysical studies at the atomic scale, thus enabling us to reach our current level of understanding and adapt them to various applications.

The bacterial flagellum is a complex and dynamic nanomachine that propels bacteria through liquids. It is composed of a basal body, a hook, and a long filament.

Flagellar assembly is a complex and energetically costly process, triggered by environmental stimuli and, consequently, finely regulated at the transcriptional, translational, and post-translational levels. Besides its role in locomotion, the filament is crucial for several other aspects of bacterial survival, reproduction, and pathogenicity, such as surface adhesion, the secretion of virulence factors, and biofilm formation.

W. Wilson et al 2007 stated that [5], Here, an agent-based 3D model is formulated to describe the establishment of single expanding bacterial colonies by the physical force of their growth.

With a single set of parameters, the model captures key dynamic characteristics of colony growth by nonmotile, non-EPS-producing E. coli cells on hard agar. The model, supported by experiments on colony growth in different nutrient types and concentrations, suggests that the radial expansion of colonies is not nutrient-limited as commonly believed, but by mechanical forces.

We announce again that the subject of this article is how the macroscopic growth of the bacterial colony follows the mechanical forces of the electric and magnetic fields in agreement with W. Wilson.

"Xiaowei Zhao finds that [6]The flagellum is one of the most sophisticated self-assembling molecular machines in bacteria, Propelled by the proton motive force, the flagellum rotates rapidly clockwise or counterclockwise, which ultimately controls the mobility and behavior of bacteria.

Wolfram Siede 2018 [7] performed a groundbreaking experiment to study the microscopic and macroscopic growth of bacterial colony in a multi-hole perforated plate Fig 2. He stated that, based on cutting holes in standard medium agar plates where mature mycelium is cultured "In terms of ease, time required and spore yield".

We have found this method to be superior to others.

It is worth mentioning that we carried out our experimental techniques on a macroscopic bacterial colony with a single large circular hole in the center of the agar plate.

**ii- On the other hand**, our more recent research, although it has rarely been discussed, constitutes the macroscopic approach which is the subject of this article [2,3,8,9].

This macroscopic descriptive and quantitative approach differs considerably from the microbiological approach. Indeed, the traditional approach focuses on cellular mechanisms, such as flagellar movement and cell replication, that drive growth, while the macroscopic approach, at the macroscopic scale, explores how physical fields, such as heat and electromagnetism, influence diffusion gradients as well as the collective behavior and spatial organization of entire bacterial colonies.

But how does this new modern macroscopic model differ from the traditional microbiological model?

In summary, our research is based on a mathematical physics perspective on bacterial growth, demonstrating that the growth and organization of a bacterial colony can be accurately described by the same equations that model physical phenomena such as diffusion and electromagnetism.

mathematical physics principles, particularly are those governing diffusion and electromagnetism. Their work suggests that

bacterial growth patterns can be modeled by partial differential equations, similar to those used to describe heat diffusion.

### Key findings from their research include:

### **Diffusion equation model:**

In previous papers, the authors presented experimental evidence that the exponential growth of a bacterial colony on a 2D surface precisely follows the solution of the heat diffusion equation, including its source/sink term and boundary conditions [2,3,9,10,11].

### **Electromagnetic Fields and Pattern Formation:**

Our studies show that the growth and surface movements of a bacterial colony are influenced by its intrinsic electric and magnetic fields. These electromagnetic fields, according to our study, help explain the complex patterns that form within colonies during macroscopic bacterial growth.

### **Magnetotastic Bacteria:**

In particular, we studied how magnetotactic bacteria, which contain iron-rich compounds, react to these intrinsic electromagnetic fields. We observed that these bacteria can separate from the rest of the colony and follow the magnetic field lines in concentric circles.

### **Bacterial "walking":**

The authors suggest that the electric and magnetic fields produced by the bacterial colony constitute an important driving force for bacterial movement, which we describe as "walking." This idea challenges the classical view that flagella and pili are the only means of locomotion.

Traditional microbiology explains macroscopic bacterial growth through individual cell multiplication and its interactions with the environment at the microscopic scale. While this explanation is accurate, our work is more general and focuses on the macroscopic scale. It suggests that the collective behavior of millions of bacteria within a colony can be described by the principles of mathematical physics, and more specifically:

Unlike the traditional microscopic approach, which focuses on the cellular mechanisms (flagellate movement and cell replication) underlying growth,

our research explores how physical fields, such as electromagnetism and diffusion gradients, influence the collective behavior and spatial organization of bacterial colonies.

In summary, this research offers a mathematical physics perspective on bacterial growth, demonstrating that the growth and organization of a bacterial colony can be accurately described by the same equations that model physical phenomena like diffusion and electromagnetism.

To avoid dwelling too much on the details of the introduction, let's move directly to Section II, which is devoted to the theory.

## II. Theory

This theory is not yet fully developed physically and biochemically, but it is reliable and destined to prevail sooner or later.

It is worth noting that this is the first time the modern theory of bacterial locomotion has been linked to its domain of action.

Recently, in 2020, a new breakthrough statistical theory in 4D unitary x-t space based on B-Transition matrix chains which is a product of the theory of Cairo techniques emerged [14,15].

It is an advanced, concrete theory of artificial intelligence called the Cairo Techniques Theory.

In this article, the theory, which models the macroscopic behavior of bacteria in nature, has proven capable of resolving the spatiotemporal evolution of aeropoietic bacteria in the most general cases, as demonstrated by experiments and pure mathematical physics.

### In conclusion, we can hypothesize the following:

The same theory can explain the spatiotemporal evolution of aerobic bacteria in most general cases.

We therefore currently have two distinct theories on the mechanics of bacterial movement: one based on the original classical biochemical theory, and the other on a mathematical physics approach to bacterial growth, supplemented by concepts from advanced biochemistry.

This article addresses this topic as shown in figures 3,4,5.

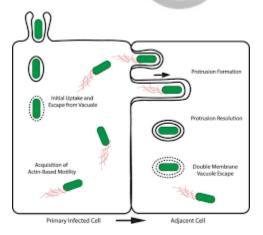
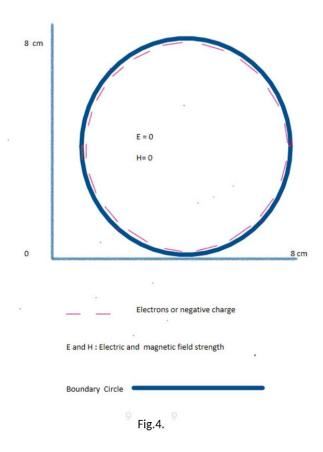
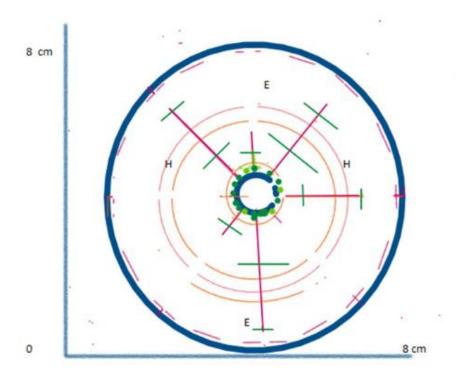


Fig.3..Bacteria walk mechanism[1]





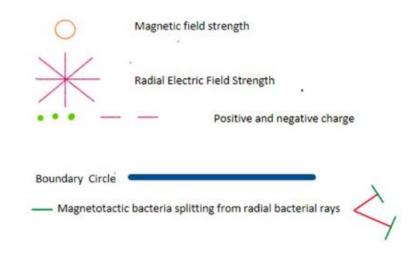


Fig.5.

In Fig.4.The colony of HOLLOW Agar bacteria zero intrinsic E and H fields.

## **III-Experimental setup and results**



Fig 5. Fully developed aerobic bacterial colony :x1.

Note also that the starting radial rays of the field E are not yet complete and that the tiny elements of the circular field **H** are in their first phase of formation perpendicular to E (**H.**⊥.**E**).

In the case of the early phase where the number and E densities are low, no complete radial or circular pattern is observed.

But both radial (E-Field driven) and circular (H-Field driven) beams progress at the same time although radial beams are much faster.

This instantly leads us to an alternative hypothesis that *nature* itself equips every bacterial cell with one or more free electrons as a "walking" mechanism to be able to survive and search for food.

The bacterial cell carries a negative charge Q= N e on its surface, (where N is the number of free electrons and e is the electronic charge equal to-1.6 E- 19 Coulomb).

However, we will not discuss in detail the origin or the mechanism of generation of negative charges in different types of bacterial cells in order to focus on the mechanism of its transport movement.

For this, we propose the following hypotheses:

i-The so-called legs are actually negatively charged appendages that extend radially and act like strings or ropes pulling the bacterial cells in a direction anti-parallel to its own intrinsic E and at the same time they influence the bacteria rich in iron via magnetic force, i.e. in circular motion normal to E.

ii- The higher the number of free electrons in a particular bacterial species, the greater its negative charge on its surface. This may be a function of the number of evolutionary stages it has passed through

However, we will not discuss in detail the origin or the mechanism of generation of negative charges in different types of bacterial cells

And therefore inhibits its ability to survive and adapt to food scarcity conditions over time in 2D and 3D geometric space.

iii-The bacterial cell has one or more free electrons and the bacterial colony contains millions of cells which produce a radial electric field and a circular magnetic field according to the EMW theory.

It is therefore the electromagnetic field of the macroscopic bacterial colony that is responsible for the macroscopic movement of bacteria passing through the holes without food.

This particular hole-crossing move is mistakenly called Leg March.

The experiment of Fig.2, Fig.5 and Fig.6 shows that the plane or surface macroscopic growth of a bacterial colony in the case of two concentric food rings follows two directions,

i-its own electric field in the radial direction i.e. along its pseudolegs and also

ii - along its own magnetic field in concentric circles, i.e. perpendicular to its pseudo-legs.

But why do bacterial cells move or walk so slowly, taking days or even weeks to bridge a gap of a few centimeters?

The answer is that the bacterial cell contains one or a few N of free electrons and the electrostatic charge (Nxe where e = -1.6 E-19 C) force Fes divided by the heavy mass of the bacterial cell is extremely low.

A similar analysis shows that the magnetic force Fm is still much less,

$$Fm = Ne Vd x B , , , , (2)$$

Fm/Fe= µ Vd

Since Vd the drift speed rate is practically zero, the Fm only appears for bacteria containing ferrous compounds ( $\mu$  iron/ $\mu$  water >1000)Which may be the reason why the bacterium accelerates faster in the radial direction and its radial structure is completed before its circular structure as shown in Figs 5,6

## III. Experimental Results

Two sets of experiments were performed on the macroscopic growth of bacterial colonies, namely,7 days Growth of bacteria on flat food surface (high quality Egyptian bread) maintained at 4-5

degrees Celsius with pH 7, normal air and humidity [1,8,9]Food Agar has the shape of two concentric rings.

Inner ring radius R1 = 1.5 cm, outer ring R2 = 4.5 cm, and iron-rich food agar content 0.3 mg/1 g.

Figure 5 shows the late developed dense phase of bacterial colony experiments.

It also shows the well-developed complete radial and circular patterns of the hollow bacterial colony.

The same experimental conditions as this case except that the macroscopic growth of bacteria was observed in its early phase after four days.

Results of this experiment is presented on Fig 6 where radial rays of the field E are not yet complete and that the tiny elements of the circular field **H** are in their first phase of formation perpendicular to **E**.

Details of the experiments (a,b) and experimental setup are also explained n ref. 2,3,8,9.

It should be noted that:

- i- The agar plate should not be compact but hollow, with appropriate R1/R2 ratios for optimal results.
- ii- The agar is rich in iron compounds and/or iron oxide (magnetite Fe3O4) [more than 0.1 mg/g].
- iii- The temperature of high-quality Egyptian bread should be maintained between 4 and 5°C for stable bacterial colony growth.

### IV. Conclusion

The "walking mechanism" of bacterial cells and the role of their legs and falagellae in their transfer or walking are studied in depth

Additionally, the structure of bacterial cells and the role of their own thermal diffusion field, electric and magnetic fields constructed by the geometric Dirichlet boundary conditions in their transfer or walking mechanism have been thoroughly discussed.

Experimental study and theoretical analysis show that the bacterial cell colony is negatively charged due to the presence of free electrons which produce an electric field flowing radially outward from the colony and a circulating magnetic field surrounding the colony.

Bacterial cells can move radially outward and cross foodless spaces through the mechanism of forcing the bacterial colony's own electric fields in addition to the swimming or crawling motion of the bacteria's legs and flagella.

In summary, all types of bacterial cells possess one or more free electrons, which compels them to move along the radial and circular lines of their own free magnetic and electronic fields. This is why macroscopic bacterial growth can extend into areas devoid of nutrients.

ii- The higher the number of electrons in a given species of bacteria, the greater its negative free charge on its surface and the greater its ability to survive or adapt to conditions of food scarcity.

NB1. All experiments in this article were produced using the author's laboratory.

NB2. The author uses his own double precision algorithm, such as that of references 16,17,18.

#### No ready-to-use Python or MATLAB algorithms are needed.

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