Networks of Bacteria Colonies: A New Framework for Reliable Molecular Communication Networking

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Abstract

The prospect of new biological and industrial applications that require communication in micro-scale, encourages research on the design of bio-compatible communication networks using networking primitives already available in nature. One of the most promising candidates for constructing such networks is to adapt and engineer specific types of bacteria that are capable of sensing, actuation, and above all, communication with each other. In this paper, we present networking of bacteria colonies via molecular communication in a diffusion channel. The node in the network itself is a population of bacteria; as an individual bacterium (biological agent) is a tiny unreliable element with limited capabilities. Each single bacterium in the population senses a parameter of interest (e.g., a chemical) and reacts accordingly. The overall response of the bacteria population in a node is transferred to the next node in the network and sensed by its bacteria. We model the transmission and reception process of each individual bacteria through which we obtain the overall functionality of a node. Two scenarios are discussed as examples. Specifically, we will consider a line network formed by these nodes. We will study transfer of information and also present some results on the practical signaling, reliability, and latency in such a multihop line network. We will also study the consensus problem in a uniformly distributed network and show how the nodes can reach a common estimate for a particular parameter in the environment.

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I. INTRODUCTION

Emergence of new applications in biomedicine, e.g., smart drug administration and monitoring systems as well as several prospective industrial and security related applications including surveillance for biological and chemical attacks require network designs that are 1) capable of operation in micro-scale 2) bio-compatible, 3) cheap to deploy, 4) low energy and can harvest energy from the environment, and possibly 5) self-organized. Such criteria necessitate the development of alternative networking paradigms. One of the most promising candidates for constructing networks that satisfy the requirements mentioned above is adaptation and engineering of specific types of bacteria that are capable of sensing, computation, actuation, and above all, communication with each other [5], [6]. The existence of a form of communication using molecules that occurs naturally among bacteria has been confirmed. Communication enables single cells to gather and process sensory information about their environment and evaluate and react to chemical stimuli. Single cells use special types of molecules for sensing their environment or sending information to the outside world [6].

One phenomenon that in particular demonstrates the essential components of communication among cells is “Quorum Sensing” [6]–[9]. Quorum Sensing can be viewed as a decentralized coordination process which allows bacteria to estimate the density of their population and regulate their behavior accordingly. To estimate the local population density, bacteria release specific signaling molecules. These molecules are subject to diffusion process [6], [10], [11]. These signal molecules will then reach the neighboring bacteria providing them with information about other bacteria in the environment. As the local density of bacteria increases, so will the concentration of the molecules in the medium. Collaboratively, bacteria use quorum sensing to coordinate actions (mostly energy expensive) that cannot be carried out by a single bacterium. This process, captures most of the important components of a molecular communication system in micro-scale.

Arguably, the most dominating form of communication at the scale of microorganisms, as observed in communication among bacteria, is molecular communication, and in particular, Diffusion based Molecular Communication (DbMC) [1], [2], [12], [13]. The essence of DbMC is embedding the information in the alteration of the concentration of the molecules and relying on diffusion to transfer the information. Hence, the behavior of a single molecule which exhibits so much randomness is of no importance. Understanding primitives of information processing and communication in bacteria would enable us to design networks with far more capabilities in their

domain.

Molecular communication and nano-networks have been studied in the literature with different perspectives. In [14] and [15], fundamentals of molecular communication in nano-networks have been discussed. They include the channel description, channel capacity discussion, protocol design and experimental validation setup. In [16], several challenges that differentiate molecular communication with conventional wireless communication have been studied. In [17], [18] and [19], the capacity of molecular communication with and without drift are discussed. A framework to study such systems are introduced in [20] and in [21] theoretical models are introduced for the molecular communication networking. In [22], the behavior of a reaction network of interacting molecules is discussed. In our previous works, we studied some of the relevant molecular communication problems, especially, the impact of diffusion channel [1] and molecular receptors [2] on the achievable rates of communication between a source and a destination. In this paper, inspired from the natural molecular communication and collective sensing occurring in bacteria, we investigate the Molecular Communication Networking (MCN). MCN utilizes the primitives of molecular communication to engineer a network of bacterial colonies capable of data gathering and relaying.

In contrast to the previous vision that the molecular communication incurs between two entities (e.g., two bacteria); here, we focus on an architecture in which a cluster of biological entities (i.e. a cluster of bacteria) communicate with another cluster. We will refer to each of these clusters that are trapped in a chamber as a biological *node*. The basic building block of the network are genetically modified bacteria to transfer information from one point to another or alternatively implement a sensor network. These biological entities are very primitive and unreliable, and hence, incapable of providing reliable communication and sensing by themselves. However, when a cluster of these biological agents form a biological node, they are collectively capable of reliable transmission and reception of information [23], [24]. The information to be transferred is acquired from monitoring some features of the environment by the nodes. The information may be binary such as whether or not a specific chemical substance is present in the environment or it can be the quantity of a chemical substance or even temperature in the surrounding environment. The interplay of these individual agents that form a biological node capable of collective sensing and relaying information is also studied in this paper.

The contributions of this paper can be summarized as follows:

- Introducing a new framework for molecular communication.
- Designing nodes in the network using engineered bacteria.
- Enabling reliable communication in the network out of unreliable bacteria.
- Introducing molecular signaling schemes for communication in the network.

The rest of the paper is organized as follows. In Sec. II, we describe the functionality of MCN and its components. In Sec. III, we describe the functionality of the nodes in MCN. The reliability and signaling schemes are discussed in Sec. IV. In Sec. V, we describe the relaying scenario in MCN and discuss the latency and reliability problems and in Sec. VI, consensus problem in MCN is discussed. Finally, Sec. VII concludes the paper.

II. THE BUILDING BLOCKS OF MOLECULAR COMMUNICATION NETWORKS

In this section, we describe the configuration and constituent modules of MCN. The schematic for a general MCN is shown in Fig. 1. The basic elements of MCN are engineered bacteria. The placement of the bacteria and their orchestrated behavior play a key role in forming the network. In Fig. 1(a) we have depicted a hypothetical network where a number of nodes (to be defined later) on the left perform a form of distributed processing whose result is then relayed to a sink node on the right. The very basic questions that arise are 1) what constitutes a node, and 2) how a message is transmitted between the nodes.

Although a single biological agent is capable of those tasks by itself, it would be very challenging (if not impossible) to form a reliable network out of these agents each acting as a single node (such as a single node in sensor networks). The rationale is that a single bacterium is too primitive to carry the transmission and reception tasks with high reliability. It is more reasonable to cluster these agents and orchestrate the cluster activity to perform these sophisticated tasks (e.g., sensing/reception and transmission) [23]. Therefore, we consider each node in the network to be comprised of genetically engineered bacteria that are contained in a chamber, depicted in Fig. 1(b). One realization of these nodes are the trapping chambers implemented on a chip [25], [26]. All the bacteria in a node are able to sense the same concentration of molecules inside the chamber and the individual molecule production of each bacterium affects the concentration of molecules in the environment; i.e., the overall response of molecule production of several bacteria will be the superposition of individual responses. This mechanism enables bacteria to collaboratively produce high concentration of molecules that can travel and be sensed from

longer distances.

The notion of the node introduced above motivates investigation of two modes of communication in the network: *intra-node* and *inter-node communication*. The intra-node communication of a node occurs between the chamber and the bacteria residing in it or vice versa. The objective of the intra-node communication is to coordinate the molecule production of bacteria in a node or transfer the information from the bacteria to the chamber circuitry. On the other hand, inter-node communication is used for transferring the information from one node to another which can be farther apart. Due to the distance between nodes, the concentration of molecules needed for the inter-node is much higher than the concentration used in intra-node communication. Hence, the aggregate response of bacteria is needed in this phase. It is important to note that intra-node and inter-node communications can be decoupled by considering a different type of molecule for each communication. We call the molecules used in the intra-node as type I and the ones used in inter-node communication as type II molecules. The idea of using multiple molecules for the molecular communication is inspired by the existence of quorum sensing with multiple molecule types in some bacteria families such as *Vibrio Harveyi*. This bacterium is believed to use multiple molecule types for quorum sensing; one type can only be detected by *Vibrio Harveyi* bacteria while others can be detected by the entire *Vibrio* family [27].

In short, functionalities of a MCN network for transferring the sensed information to other parts of network are as follows. First, the bacteria inside a node sense the environment and inform the chamber by the production of light or GFP (Green Fluorescent Protein). The chamber receives and decodes the information and, with an appropriate timing, produces type I molecules to activate the bacteria in the node. In the second phase of communication, due to reception of type I molecules, bacteria start to emit the type II molecules which would reach the neighboring nodes in the network. These two modes of communication will be discussed in details later. In the next section, we will discuss the building blocks of the MCN in detail.

III. NODE MODEL IN MCN

Here, we study the characteristic of the nodes. Each node is comprised of a chamber and n bacteria residing in it. Nodes can be considered as an independent entity who can sense the concentration of different types of molecules, process the information embedded in the concentration of molecules and release molecules. The functionality of the node is formed by the interplay of the chamber and the bacteria inside it. Type I molecules are not able to pass

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5 through the chamber walls and are aimed for communication between the chamber and the bacteria in the node.
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7 On the other hand, type II molecules can freely diffuse outside the node and are responsible for communication
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9 between the nodes in the network. Each node can act as a transmitter or a receiver at a time. The reception or
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11 production of molecules in a node is performed by the population of bacteria inside the chamber. In order to
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13 explain the functionality of the nodes, we first introduce the characteristics of engineered bacteria inside a node
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15 and explain a model given in literature for related processes inside the cell.
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19 *A. Bacteria Functionality*

22 Bacteria are the primitive elements of MCN that are genetically engineered to perform specific tasks upon the
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24 reception of certain types of molecules. Some portions of DNA content of a bacterium can be manipulated to
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26 construct specific features like signal transmission and/or reception. One common technique involves manipulation
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28 of DNA molecules within bacteria that are separate from the chromosomal DNA called *plasmid* [28], [29], as
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30 shown in Fig. 1(c). A plasmid is a double-stranded DNA molecule that can be viewed as an ordering of genes
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32 each responsible for a function. For example, deactivating the receptor (transmitter) genes in some variants of
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34 bacteria can provide transmitter (receiver) bacteria.
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37 Bacteria can also work as transceivers, i.e., they sense the concentration of molecules in the medium and release
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39 molecules back into the medium. More sophisticated functionalities can also be implemented through the same
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41 process. For example, some bacteria can be engineered to be able to receive specific types of molecules and
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43 perform particular logical operations upon reception [5].
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46 Here, we focus on the molecule reception (sensing) process of bacteria. In the following sections, we explain
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48 how this process can be used by the nodes in MCN to design transmitters and receivers. Assume each bacterium
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50 has N receptors for a specific molecule type, for example, the U-shaped receptors in Fig. 1(c). Further, we assume
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52 that the receptors act independently. As in [1], [2], assume the information is embedded in the concentration of
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54 the molecules. We consider the steady-state behavior of the entrapment of molecules by the receptors and ignore
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56 the transient behavior. Activation of a receptor (i.e., entrapment of molecules in the receptor) triggers a chain of
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58 processes that results in the production of fluorescent or a specific molecule type by the bacteria. The entrapment
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60 of molecules by the receptors has a probabilistic nature in a way that more concentration of molecules results in
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higher probability of the entrapment of molecules in a receptor and hence, higher probability of activation.

In [10], a set of differential equations are provided that account for the quorum sensing process in bacteria. In particular, these equations account for the reception of molecules, protein complex formation, transcription of genes and performing a specific task such as production of fluorescent or a specific molecule type. The reception of molecules has a probabilistic nature such that increasing the concentration of molecules results in the higher probability p of molecule binding in the cell. The differential equation accounting for probability of binding of molecules to the cell receptors is given by [10]:

$$\dot{p} = -\kappa p + A\gamma(1 - p), \quad (1)$$

where A is the concentration of molecules, γ is a constant for the input gain and κ is the dissociation rate of trapped molecules in the cell receptors. Here, $\dot{p}$ is the derivative of p with respect to time. In this model, each cell receptor (i.e., the ligand receptor) is activated with a probability that depends on the concentration of molecules in the medium surrounding the cell. This probability starts to increase from the moment a constant concentration is applied until it takes its final steady-state value p^* given by

$$p^* = \frac{A\gamma}{A\gamma + \kappa}. \quad (2)$$

As we see in (2), the steady-state probability of binding of molecules to cell receptors is an increasing function of the concentration of molecules. The process of the production of complex molecules and transcription of genes and the process of then production of fluorescent are modeled similarly as

$$\begin{cases} \dot{S}_1 &= (bp + a) - b_1 S_1 \\ \dot{S}_2 &= a_1 S_1 - b_2 S_2 \end{cases}, \quad (3)$$

where S_1 and S_2 are two post-transcription messengers and b_i and a_i are some constants [10]. In the steady state, the final product S_2 is obtained as

$$S_2^* = \frac{a_1(bp^* + a)}{b_1 b_2} \quad (4)$$

Note that S_2^* , i.e., the amount of fluorescent, has an initial value even when $p^* = 0$ and increases linearly when p^* increases. In the following, we describe the functionality of the nodes based on the behavior of individual bacteria introduced above.

B. Node Functionality in MCN

Information in the network is decoded in the concentration of type II molecules. Assume the transmitter node has concentration A_0 of type II molecules as the information to be sent to the receiver node. The chamber of the node stimulates the population of bacteria inside it with the proper concentration of type I molecules in the intra-node communication phase. The quantity of this concentration, assumed to be uniform inside the node and obtained in [24], depends on both the structure of the transmitter bacteria (in reception of type I and production of type II) and on the channel parameters. The type I molecules causes each individual bacterium to produce type II molecules with a rate that depends on the number of its activated receptors. As discussed in bacteria model, the activation of each receptor has a probabilistic nature. Since the diffusion process of molecules is linear, the overall output of the node is the aggregated output of the individual bacteria in the population.

The number of activated receptors of bacteria in the population determines the intensity of the output. We denote by X_i the number of activated receptors of i^{th} bacterium. Hence, the total number of activated receptors in the node is $X = \sum_{i=1}^n X_i$. We assume that each bacterium has N receptors at its membrane and activation of each receptor results in production of type II molecules with the rate α . The total molecular production of the nodes depends linearly on the number of activated receptors. We discuss this further in the next section.

In the inter-node communication phase, the molecules produced by the transmitter node pass through the chamber membrane into the medium. Here, the channel is the medium in which type II molecules can freely diffuse and reach the receiver node. We assume that the size of the nodes is negligible with respect to the distance between them and hence, the concentration of type II is uniform inside the receiver node. The steady-state response of diffusion of molecules is the parameter of interest sensed by the receiver node. A brief review of diffusion channel is given in the Sec. V. The overall molecular production of the transmitter bacteria passed through the channel results in the concentration of type II molecules at the receiver node which has average value of A_0 .

At the receiver node, the molecules pass through the chamber and stimulate the bacteria residing in the receiver node. Upon the reception of molecules, each individual bacterium goes through a chain of processes to start the intra-node communication phase in the receiver. This chain of processes is similar to the one in transmitter bacteria but the receptors and the protein that type II molecules would bind to are different. Moreover, the output of the reception of type II molecules is different from the output of reception of type I ones. The output of receiver bacteria

can be sensed by the chamber and can be in different forms by programming the plasmid inside the bacteria. Here, we assume that the output of reception of type II molecules is fluorescent. The aggregate fluorescent production of bacteria is sensed by the chamber in order to infer the information about the concentration of molecules.

In the next section, we will look closely into the process explained above and discuss the noise in the different stages of communication between the transmitter and receiver. This will allow us to study the reliability of communication and signaling schemes that will be achievable by using such nodes in the network.

IV. RELIABILITY IN MCN

In this section, we discuss the reliability and probability of error in the communication between two nodes in MCN. Then, we will introduce signaling schemes that can achieve particular reliabilities. There are three major sources of noise in communication between the transmitter and the receiver. First is the molecule production inaccuracy of the bacteria inside the transmitter node. As explained in the previous section, the individual behavior of bacteria inside the population is probabilistic. Moreover, the parameters of the probability distribution of their outputs are different as well. This would translate into variance in the aggregate output of the transmitter node. As shown in [24], the probability of activation of a receptor of a bacterium in the population can be written as

$$p = p^* + p^*(1 - p^*) \frac{\epsilon_\gamma}{\gamma} - p^*(1 - p^*) \frac{\epsilon_\kappa}{\kappa}. \quad (5)$$

where $p^* = \frac{A\gamma}{A\gamma + \kappa}$ is the noiseless probability and ϵ_γ and ϵ_κ are zero-mean normal noises with variances σ_γ^2 and σ_κ^2 , respectively, accounting for the discrepancy among bacteria parameters. The average and variance of X , the number of activated receptors, are given by [24]

$$\begin{cases} \mathbf{E}[X] &= nNp^* = nN \frac{A\gamma}{A\gamma + \kappa} \\ \text{Var}[X] &= nN^2p^{*2}(1 - p^*)^2 \left(\frac{\sigma_\gamma^2}{\gamma^2} + \frac{\sigma_\kappa^2}{\kappa^2} \right), \end{cases} \quad (6)$$

where N is the number of receptors in a single bacterium and n is the number of bacteria in a node. As we observe from (6), increasing n results in larger $\frac{\mathbf{E}[X]}{\sqrt{\text{Var}[X]}}$ which translates into more precision in the output of the transmitter node. Hence, by using larger number of bacteria inside the chamber, not only we can produce wider range of outputs (i.e., the more spread concentration of molecules at the receiver) by the nodes, but also the outputs can be made more differentiable. Note that as we see in (6), the variance of the output decreases and hence, the precision increases when p^* is closer to either 1 or 0.

The second source of the noise is the diffusion channel. In practice, the free diffusion may not be possible and obstacles or interference with other types of molecules can affect the communication inside the diffusion channel. On the other hand, the uncertainty about the channel parameters by the transmitter is another factor to consider in the communication. In order for the transmitter to send the proper concentration of molecules to the receiver-by stimulating the bacteria with proper concentration of type I molecules- the transmitter must know exactly how far the receiver node is located and what the diffusion coefficient of the medium is. Uncertainty in both parameters results in less precision of the output at the receiver.

Finally, the third source of the noise is the uncertainty in the fluorescent production of the receiver bacteria. Due to presence of type II molecules at the receiver, the receptors of each individual bacterium are activated in a probabilistic manner similar to the transmitter case. Hence, the aggregated output of bacteria contains some degrees of uncertainty which again can be reduced by using larger number of bacteria inside the node. Note that the noise introduced from each stage is carried to the next one and affects the final output which is in the form of fluorescent. We have analyzed the noise level of output in [24] and obtained the amount of information that can be inferred about the input concentration from the receiver output. The mathematical analysis of noise in the communication is discussed in [23], [24]. Note that this analysis is based on the assumption that the chambers behave perfectly or the uncertainty it introduces in producing the stimulant or sensing the fluorescent is negligible compared to the uncertainty in the performance of the bacteria.

In the following, using the nodes we described, we introduce two signaling schemes that can be used in MCN. In the first one, we modulate the information in different levels of type II molecules. Note that this modulation is possible because we use a population of bacteria inside the nodes and they are able to produce and sense different levels of concentrations. In the second scheme which we will briefly explain, different kinds of type II molecules are used in order to increase the reliability in the network.

A. *M-ary Signaling Modulation and Reliability in MCN*

In practice, we may use only a finite discrete number of levels of molecular concentrations A_0 for communication which results in discrete levels of activation probability of bacteria. We denote by A_{max} the maximum concentration of type II molecules that can be produced by a node. This maps to $p_{max} = \frac{A_{max}\gamma}{A_{max}\gamma + \kappa}$ from (2). Note that the

probability of activation of receptors can be used as the input instead of the concentration itself. In this section, we consider a specific signaling method (i.e., M-ary modulation) and study the the information exchange rate and the corresponding achieved error rate in the communication between a transmitter and receiver in MCN. Then the results can be extended for the relaying of information from the source to the destination in a line network. The range of the input is determined by p_{max} . Two factors influence the signaling performance: the number of levels of concentration and the choices for the values of those levels.

We consider the scenario in which m symbols to be chosen with uniform spacing from the interval $[0 \ p_{max}]$. The i^{th} symbol level would correspond to $p_{max} \frac{i}{m-1}, 0 \leq i \leq m-1$. We show by $p_{e,i}$ the probability of error in the detection of i^{th} symbol. Hence, the total probability of error is equal to $p_e = \sum_{i=0}^{m-1} w_i p_{e,i}$, where the weights w_i associated with the m symbols must be obtained .

We consider a hard decision scenario in decoding the symbols. In other words, the receiver chooses the closest symbol to the received one. Hence, the error occurs when the detected symbol passes the half way from the previous or the next symbol. As observed in (6), the variance of the noise, and hence $p_{e,i}$ depends on the chosen symbol i . Therefore, we have

$$p_{e,i} = 1 - \Pr \left[\frac{-p_{max}}{2(m-1)} \leq \epsilon_i \leq \frac{p_{max}}{2(m-1)} \right], \quad (7)$$

where ϵ_i comes from a $\mathcal{N}(0, \sigma_i^2)$. Here, σ_i^2 can be computed by replacing p^* with $\frac{i}{m-1} p_{max}$ in (6). As discussed in the previous section, the noise variance is the smallest when the input is closest to 0 or 1. Hence, it is intuitive to choose larger weights for the inputs closer to these two points. In our scheme, we use the weights from the optimal distribution calculated by the Blahut-Arimoto algorithm. In Fig. 2, we have shown the rate of information for different M-ary modulations versus the power of the transmitter. In this setup, we have chosen $N = 50$ and $\frac{\sigma_\gamma^2}{\gamma^2} + \frac{\sigma_\kappa^2}{\kappa^2} = 0.1$. In addition, the number of bacteria in a node is chosen to be $n = 100$. As shown by the plot, reliable communication (i.e., $p_e = 10^{-6}$) is feasible for $M = 2, 4, 8, 16$ and the required power is shown as well. For larger number of symbols, reliable communication is not possible as for the case of $M = 32$, the least error rate (by maximizing the p_{max}) would be 10^{-2} . In such cases, smaller error rates can be achieved by increasing either n or N (or both).

B. QAM Modulation

The other method that can be used to increase the amount of sent information is to use Quadrature Amplitude Modulation (QAM). In this method, multiple types of molecules are used for communication between nodes. As shown in [27], some types of bacteria are able to respond to different types of molecules in similar ways. The intensity of the output in response to these types can be different as shown in [27]. Therefore, in engineering bacteria, we can program the plasmid in order to make the bacteria responsive to specific types of molecules. Moreover, the output due to the reception of these molecules can be set to be the same or not.

For example, we may assume that the bacteria are able to produce and send only two kinds of type II molecules. For the sake of discussion, we further assume that only two levels of molecule types II-A and II-B, low and high, are produced. The outputs of the two types are assumed to be in the same form, i.e. fluorescent. Hence, four different scenarios can occur in each transmission. However, since the outputs of two types are not assumed to be differentiable, only three levels of outputs are possible which result in maximum $\log(3)$ bits of information.

On the other hand, this method can be used to diversify the communication by sending the same information via two types of molecules. It can decrease the probability of error by using a repetition error coding. As we have shown in the previous section, the error for sending the “0” bit, corresponding to low concentration, is practically zero. The error in sending the bit “1”, corresponding to high concentration, can be lowered using this modulation. In this scenario, the receiver node infers the received bit to be “1” if at least one of the molecules has been received. Hence, the error happens when both means of communication fail.

V. RELAYING THE INFORMATION IN MCN

In our setup, it is clear that the nodes can be considered as independent and relatively smart entities in a network and can use communication in the network for various reasons. We consider a relaying scenario where information is transferred from a source node to a destination node several hops away in a line network. The relaying problem is in particular of importance because of the very low communication range in MCN. In other words, nodes can only directly communicate with each other in short distances. Therefore, for applications that require transfer of information for long distances, we should resort to multihop communication strategy. In the following, we first review the channel model we use in MCN and then analyze the relaying problem in a line network. We focus on

a line network so that we do not deal with the routing problem needed in a general network.

A. Diffusion Channel Model

The molecules produced by the transmitter travel through the channel via the diffusion process. The first step is to characterize the temporal and spatial variations of molecules in the channel which follows the general diffusion equations. According to Fick's second law of diffusion the concentration of molecules $A(r, t)$ at position r at time t is computed as follows [30]:

$$\frac{\partial A(r, t)}{\partial t} = D \nabla^2 A(r, t), \quad (8)$$

Here, r is the distance of any point in the environment from the source and D is the diffusion coefficient of the medium. The impulse response of (8), is the Green's function $g_d(r, t)$ whose expression is as follows:

$$g_d(r, t) = \frac{1}{(4\pi Dt)^{\frac{3}{2}}} \exp\left(-\frac{r^2}{4Dt}\right). \quad (9)$$

This impulse response is given for the 3-D medium using the observation that 3-D diffusion is equivalent to 3 separate (simultaneous) 1-D diffusions. Since the diffusion equation is a linear equation, the solution to (8) for an arbitrary input $F(r, t)$ can be obtained using

$$A(r, t) = g_d(r, t) \otimes F(r, t), \quad (10)$$

where $\otimes$ denotes a multi-dimensional convolution operation on r and t .

In our setup, we assume that there is only one transmitter emitting molecules. Therefore, in (10), we have $F(r, t) = f(t)\delta(r)$, where $f(t)$ is the time-dependent input signal. Moreover, the output is assumed to be a pulse of length t_0 and amplitude α . In other words, the transmitter produced α molecules per second for $0 \leq t \leq t_0$. Hence, we will have

$$\begin{aligned} A(r, t) &= \int_0^t \alpha \frac{1}{4\pi D\tau} \exp\left(-\frac{r^2}{4D\tau}\right) d\tau \\ &= \frac{\alpha}{4\pi Dr} \operatorname{erfc}\left(\frac{r}{(4Dt)^{\frac{3}{2}}}\right), \end{aligned} \quad (11)$$

where $\operatorname{erfc}(x)$ is the error function complement. $1 - \operatorname{erf}(x)$, and $\operatorname{erf}(x)$ is the error function, defined by the integral

$$\operatorname{erf}(x) = \frac{2}{\sqrt{\pi}} \int_0^x e^{-u^2} du. \quad (12)$$

If the duration of pulse t_0 is long enough, the concentration at the steady-state will be

$$A^*(r) = \frac{\alpha}{4\pi Dr} \quad (13)$$

This response is valid for open free media in which the only boundary conditions are at the transmitter. If the dimension of the receiver node a is comparable to the distance r between the nodes, a factor $(1 - \frac{a}{r})$ should be multiplied to the expression in (13). In this paper, we assume that the distance between the nodes is significantly larger than the size of the nodes; hence, we ignore this small effect on the steady-state response.

As discussed in the previous section, each activated receptor contributes an amount of α to the output rate. Since the diffusion channel is linear, the total response at the receiver will be the superposition of individual responses. Hence, the steady-state concentration A_r at the receiver R will be

$$A_r = \frac{\alpha X}{4\pi Dr}, \quad (14)$$

where r is the distance between the transmitter and the receiver nodes and X is the total number of activated receptors. In the analysis above, we did not include any noise or interference in the channel and assumed that the diffusion can be described by the Fick's law. As noted in the previous section, if the parameters D or r are not exactly known to the transmitter, the response in (14) contains some degrees of uncertainty which adds to the uncertainty in the receiver output.

B. relaying reliability in MCN

Consider node 1 shown in Fig. 3. Node 1 has some information that needs to be transmitted to node $h + 1$. This information is an estimate of a local parameter that has been obtained via the bacteria at node 1, i.e., the source node. We assume that nodes 1 and $h + 1$ are located outside the effective communication range, and hence, they should use other nodes as relays to transfer information. Suppose that the effective range of each node includes only the immediate neighbors of it. The interference of molecules from the previous hops is ignored in this discussion but can be studied as a future work.

In a general network, it remains as an open research problem as to how one can perform the path discovery in a molecular communication while maintaining the realistic assumptions on the biological nodes (e.g., limited number of molecules to be released, lack of even simple addressing method, etc.). To avoid path discovery issues,

we envision that all paths from the nodes to the sink node can be preformed for the sensor network applications. This is realistic as the data is collected from the nodes to the fixed sink node and hence, the path from each node to the sink can be formed by the proper placement of the nodes. Specifically, to form a path (as in a routing path in the traditional networks), we assume that every node shuts down after relaying the information to the next node. This way, the transmitter node will not receive the molecules that are produced by the next node in the path. Hence, the information does not move back and forth the path.

Note that the processes of production of molecules, diffusion in the channel, and the sensing impose significant delay, which is in order of several hours. Further, the lingering of the molecules in the medium (before they are carried away) makes the channel to have *memory*, which in turn causes inter symbol interference [1]. Hence, in the molecular communication, we try to transmit as many bits of information as possible in one use of channel. Moreover, due to the significant distance of the nodes relative to the size of the bacteria, we can assume all agents are co-located at the center of the node and sense the same concentration of molecules.

There is a total number of h hops needed to transfer the information from node 1 to $h+1$. In the previous section, we explained the communication between two nodes which is one hop in the path. Assume the information to be transmitted by the first node is a level of concentration of type II molecules A_0 . This concentration is mapped to a probability $p_0 = \frac{A_0\gamma}{A_0\gamma+\kappa}$ as explained previously. From now on, we assume p_0 to be the parameter to transfer in the network. By using the analysis in previous section and results from [24], the information which is transferred to the next node (in the first hop) is in the form of

$$p_1 = p_0 + \frac{p_0(1-p_0)}{n}\epsilon, \quad (15)$$

where n is the number of bacteria inside the nodes. Moreover, ϵ is a zero-mean Gaussian noise with small variance σ^2 which is due to the discrepancy in behavior of bacteria. The probability p_1 is inferred by the second node through sensing the fluorescent output of the bacteria inside it and in turn is transferred to the next node. Note that by using a larger number of bacteria, the inference error becomes smaller. Since n is large, by ignoring the terms $\frac{\epsilon^2}{n^2}$, for the h^{th} hop we have:

$$p_h = p_0 + \frac{p_0(1-p_0)}{n} \sum_{i=1}^h \epsilon_i, \quad (16)$$

where ϵ_i 's are independent and come from a $\mathcal{N}(0, \sigma^2)$ distribution. Hence, the noise in each hop is aggregated

with the noise from the previous hops. For the destination node we have:

$$\begin{cases} \mathbf{E}[p_h] &= p_0 \\ \text{Var}[p_h] &= \left(\frac{p_0(1-p_0)}{n} \right)^2 h\sigma^2 \end{cases} \quad (17)$$

C. Time Latency of relaying in MCN

Here, we study the time it takes for the network to transfer information from the source to the destination node in a line network involving h hops. The latency in a single-hop communication between a transmitter node and a receiver is comprised of three stages: the time it takes for the transmitter bacteria to produce molecules, the time it takes for the channel to transfer those molecules to the receiver and finally the time it takes for the receiver bacteria to sense the molecules and produce output (e.g., fluorescent).

In order to study the latency at the transmitter and receiver side, we use the set of differential equations in Sec. III. Since the steady-state responses are the measured values, we calculate the time constants of transient response as a measure of latency. Moreover, we use the estimated parameters given in [10]. Note that these processes occur consecutively. Hence, the time it takes for each stage to reach steady-state directly affects the next stage and the overall delay will be comprised of all three time stages of delay. The time constant for the entrapment of molecules is given by $T_1 = \frac{1}{A\gamma+\kappa}$. Hence, for a typical value of concentration of molecules, $T_1 \simeq 5 \text{ min}$ which is larger for smaller values of A . The time constant for the production of protein and transcription of genes is $T_2 = \frac{1}{b_1} \simeq 1 \text{ hr}$. Finally, the time constant for production of fluorescent is obtained as $T_3 = \frac{1}{b_2} \simeq 10 \text{ min}$.

Hence, by assuming three times of the time-constant as the time required to reach the steady-state response, the overall delay at the transmitter node is obtained as $T_T \simeq 4 \text{ hr}$ using the values calculated above. Note that the production of proteins and transcription of genes are the dominating factors in the response delay which are not related to the concentration of molecules A . Hence, we can assume that the above answer holds for all the values of A in the range. The receiver node introduces delay through the similar set of processes and hence, the receiver delay is equal to $T_R \simeq 4 \text{ hr}$ as well.

The next factor to address is the time for the molecules to travel through the diffusion channel. We need to determine the time T_r in (11) that it takes for the response to approach the steady-state value. Using the Fick's law (8), T_r does not depend on A as shown in [30]. Therefore, $T_r \simeq \frac{r^2}{D}$ where r is the distance between the transmitter and the receiver and D is the diffusion coefficient. For typical values of $r = .5 \text{ cm}$ and $D = 10^{-5} \frac{\text{cm}^2}{\text{sec}}$,

we obtain $T_r \simeq 3.5 \text{ hr}$. Hence, the time for the molecules to travel in the channel is in the same order of magnitude as the time it takes for the transmitter and the receiver to process.

The discussion above confirms the slow nature of the molecular communication. In particular, the communication in one hop in Fig. 3 can take several hours which has to be multiplied by h to give the answer for the time needed to transfer the information over h hops. Hence, MCN should be used in the scenarios where several hours of latency introduced by the network, can be tolerated. The detection of the existence of a particular chemical in a biological environment can be among those applications.

VI. CONSENSUS PROBLEM IN MCN

As an example of another realization of the molecular network, here, we consider the consensus problem in a sensor network formed by MCN. In a general consensus problem, nodes in a network communicate with each other to obtain the best estimate given their initial estimates. The average value of these initial estimates is considered to be an important measure that can be considered as a goal in a network. Here, by producing different rates of molecules, nodes give information about their own estimates and by sensing the concentration of molecules in their proximity, they obtain information about the estimates of other nodes. The key idea here is the linear nature of diffusion process. It lets the nodes to determine the aggregation of estimates corresponding to other agents.

We assume a network of nodes that unlike the relaying scenario, each node can communicate with all the other nodes in the network. Therefore, production of molecules by a node affects the concentration around all the other nodes in the network. Note that in a special case that nodes are not very close to each other, nodes can only have effective communication with their immediate neighbors which results in the same assumption as in the relaying scenario.

We consider a network of M nodes deployed uniformly. Each node senses the environment for a particular parameter of interest. Nodes map their estimated value into a concentration of type II molecules. We assume that the initial estimates θ by nodes has a Gaussian distribution with mean μ and variance σ_0^2 . The average of initial estimates is the best unbiased estimate for μ with variance $\frac{\sigma_0^2}{M}$. Hence, the objective is to produce the average of all the estimated concentrations at the vicinity of all the nodes. In a network with sufficiently large M , each node observes the same relative distances r_{ij} to the other nodes in the network (except for the nodes on the boundary

of the network whose effects are negligible when M is large). Since the network size is large, each node has effective communications with only a specific number of nodes which is determined by the range of nodes.

The concentration of molecules in the steady-state A_i^* at the vicinity of the i -th node can be obtained by

$$A_i^* = \sum_{j=1}^M \frac{f_j}{4\pi D r_{ij}}, \quad (18)$$

where f_j is the rate of production of j^{th} node in the network. In (18) we have used the linear nature of diffusion process in order to calculate the superposition of individual responses of nodes in the network. Hence, in the vectorized form we have:

$$\mathbf{A}^* = \mathbf{R}\mathbf{f} \quad (19)$$

where $\mathbf{A}^*$ and $\mathbf{f}$ are both vectors and $\mathbf{R}$ is a symmetric matrix in which $\mathbf{R}(i, j)$ is equal to $\frac{1}{4\pi D r_{ij}}$. Since, the network is uniform, the columns and rows of $\mathbf{R}$ are permutations of each other.

We proposed an iterative algorithm in [31] that results in consensus in such networks. In this algorithm, nodes need to have the knowledge of S the sum of any column of matrix $\mathbf{R}$. In iteration l , chamber in node i stimulates the bacteria inside it to produce molecules with rate $f_i = \frac{\hat{\theta}_i(l)}{S}$. where $\hat{\theta}_i(l)$ is the estimate of node i at iteration l . The production of molecules by all the nodes result in a variant concentration of molecules at the vicinity of the nodes. These concentrations are sensed by corresponding bacteria in each node and through intra-node communication, the output of bacteria (e.g., fluorescent) is measured by their chamber.

Each node updates its estimate by $\hat{\theta}_i(l+1) = c_i^*(l)$ in response to the measured output, where $c_i^*(l)$ is the concentration at the node i in the iteration l . Note that the process of sensing the concentration, updating the estimates and sending with new rates take some time that we can assume the channel has been rest meanwhile. Hence, we will have an iterative equation as

$$\theta(l+1) = \theta(l)\tilde{\mathbf{R}} \quad (20)$$

where $\theta(l)$ is the vector estimates of the nodes and $\tilde{\mathbf{R}}$ the normalized version of $\mathbf{R}$ is a doubly stochastic matrix. In such a matrix we know there is one eigenvalue λ_1 equal to one and all the other eigenvalues are less than one [32]. Moreover, the eigenvector v_1 corresponding to the λ_1 is a uniform vector corresponding to the consensus vector.

Theorem 1. *The iterative algorithm above converges to the average value of initial estimates with a rate that depends on the second largest eigenvalue of $\tilde{\mathbf{R}}$.*

Proof: The eigenvalue decomposition of $\tilde{\mathbf{R}}$ is

$$\tilde{\mathbf{R}} = \mathbf{Q}\mathbf{\Lambda}\mathbf{Q}^{-1} = \mathbf{Q}\mathbf{\Lambda}\mathbf{Q}^T. \quad (21)$$

The estimate of nodes at iteration n is given by

$$\begin{aligned} \boldsymbol{\theta}(l) &= \tilde{\mathbf{R}}\boldsymbol{\theta}(l-1) \\ &= \tilde{\mathbf{R}}^l\boldsymbol{\theta}(0) \\ &= \mathbf{Q}\mathbf{\Lambda}^l\mathbf{Q}^T\boldsymbol{\theta}(0). \end{aligned} \quad (22)$$

Since all the eigenvalues except for $\lambda_1 = 1$ are smaller than one, all the entries in the main diagonal of matrix $\mathbf{\Lambda}$ approaches zero except for $\Lambda_{11} = 1$. Hence, we have

$$\begin{aligned} \lim_{l \rightarrow \infty} \boldsymbol{\theta}(l) &= v_1 v_1^T \boldsymbol{\theta}(0) \\ &= \frac{1}{M} \left(\sum_{i=1}^M \theta_i(0) \right) \mathbf{1} \\ &= \theta_{av} \mathbf{1}. \end{aligned} \quad (23)$$

where $\mathbf{1}$ is the vector of all ones. As we see in (23), after sufficient number of iterations, the estimate of nodes approaches to the average of initial beliefs. In order to quantify the rate of convergence, we look into the variance of estimates of nodes. As mentioned before, the smallest variance that can be achieved is $\frac{\sigma_0^2}{M}$ which corresponds to the average value. The covariance matrix $Cov(l)$ of the estimates at iteration l is given by

$$\begin{aligned} Cov(l) &= \mathbf{E} [(\boldsymbol{\theta}(l) - \mathbf{E}[\boldsymbol{\theta}(l)])(\boldsymbol{\theta}(l) - \mathbf{E}[\boldsymbol{\theta}(l)])^T] \\ &= \tilde{\mathbf{R}}^l \mathbf{E}[(\boldsymbol{\theta}(0) - \mathbf{E}[\boldsymbol{\theta}(0)]) \\ &\quad (\boldsymbol{\theta}(0) - \mathbf{E}[\boldsymbol{\theta}(0)])^T] (\tilde{\mathbf{R}}^T)^l \\ &= \tilde{\mathbf{R}}^l Cov(0) \tilde{\mathbf{R}}^l. \end{aligned} \quad (24)$$

where the fact that all powers of a symmetric matrix are also symmetric is used. Since the initial estimates are considered to be independent of each other, $Cov(0) = \sigma_0^2 \mathbf{I}_{M \times M}$ where $\mathbf{I}$ is the identity matrix. Hence, from (24),

$Cov(l) = \sigma_0^2 \tilde{\mathbf{R}}^{2l}$. We denote the diagonal elements of the matrix $Cov(l)$ by $Cov_{ii}(l)$ which gives the variance of the estimate of each node at iteration n . For large n , we consider only the effect of λ_2 , the second largest eigenvalue of $\tilde{\mathbf{R}}$. Thus, by using the decomposition in (21), we have

$$Cov_{ii}(l) = \sigma_0^2 \left(\frac{1}{M} + v_{2i}^2 \lambda_2^{2l} \right), \quad (25)$$

where v_{2i} denotes the i^{th} element of eigenvector v_2 corresponding to λ_2 . Since the norm of eigenvector v_2 is one, then $v_{2i} \leq 1$, $i \in 1, 2, \dots, M$. Hence, the variance of the estimate of node i at iteration n is obtained as

$$\sigma_i^2(l) \leq \sigma_0^2 \left(\frac{1}{M} + \lambda_2^{2l} \right) \quad i \in 1, 2, \dots, M. \quad (26)$$

■

It can be proved that when matrix $\mathbf{R}$ becomes more sparse, i.e. the columns and the rows contain more zeros, the rate of convergence falls. This can be explained by the fact that a more sparse $\mathbf{R}$ means a more extended network. Hence, more number of iterations are needed to spread the information in the network and hence, reaching consensus will be more time consuming. In particular when $\mathbf{R}$ becomes a diagonal matrix, the variance does not converge to $\frac{\sigma_0^2}{M}$. This case is equivalent to the scenario in which none of the nodes are able to communicate with each other and their initial beliefs cannot be improved.

VII. CONCLUSION

In this paper, we studied networking of bacteria colonies. Such networks are envisioned to perform tasks in biological environments where conventional networking paradigms fail to operate. We proposed that these networks can be formed via several bacteria agents acting as a whole like a node; performing collective sensing, signal reception and transmission. The functionality of the engineered bacteria used in such networks was discussed. It was also explained how these functionalities can be used to form reliable transmitter and receiver nodes. The reliability and signaling schemes were discussed in such a network as well. In addition, the relaying of information in the MCN was discussed. In particular, latency and reliability of transferring information in a multihop line network were studied. Finally, we introduced the consensus problem in such networks and explained how nodes can reach a common estimate using an iterative algorithm.

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(a) A hypothetical network of biological nodes.

(b) A biological Node.

(c) A biological agent.

Fig. 1. Configuration and constituent modules of MCN.

Fig. 2. The information rate versus the maximum power of the transmitter for different M-ary schemes

Fig. 3. Multihop information relaying

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