MODELING OF INTERCELLULAR CONNECTIONS IN THE SINOATRIAL NODE*

DANUTA MAKOWIEC

Institute of Theoretical Physics and Astrophysics, Gdańsk University
Wita Stwosza 57, 80-952 Gdańsk, Poland

(Received January 12, 2010)

Electrochemical activity of the sinoatrial node — the first natural heart pacemaker, relies on coordinated activity of sinoatrial cells and on signal transduction by intercellular connections. The modified Greenberg–Hastings automaton is used to model the electrochemical activity of a cell and basic intercellular interactions. A stochastic 2D lattice with preference set to lateral connections is a starting point in the construction of the network of intercellular connections. Then a flat structure of the network is carefully wrinkled by rewiring procedure. The rewiring is restricted to neighboring cells — local rewiring, and it favors densely connected cells — preferential rewiring. In simulations we find that if density of intercellular connections reaches $d = 0.60$ then spirals of activity clusters emerge robustly. However to observe strong spirals: oscillating with the shortest period possible and driving dynamics in the whole network, the intercellular connections have to be rewired locally and preferentially. The critical value of density corresponds very accurately to the known value at which the canine sinoatrial node works.

PACS numbers: 87.19.Hh, 87.17.Aa, 87.18.Mp

1. Introduction

A normal contraction of the heart is initiated by the sinoatrial node (SAN) — the first natural pacemaker of the heart [1–3]. The area of the SAN is well defined basically due to the anatomical boundaries. The SAN is located on the right atrium at the junction of the crista terminalis with venous tissue — the superior and inferior vena cava, and the intercaval region between the two great veins. Moreover, the SAN tissue differs significantly from the other parts of the cardiac muscle [3–7]. The myocytes from the SAN

are small and “empty” — without well-developed cardiac sacroplasmic reticulum which indicates that these cells do not have ability to contract. But in the contrast with other cardiac cells, the pacemaker cells have the ability to produce the rhythmic electrochemical changes in the cellular membrane — called the self-excitation property.

The space organization of the SAN cells is also different from organization of myocytes in the working parts of the heart. The cells are scattered rather rarely, like at random, they are surrounded by connective tissue, mainly collagen and fibroblasts. But the basic way to pass the membrane activity is by cell-to-cell connections, achieved here via gap junctions built from the connexin Cx45 while the connexin Cx43 dominates in the rest of the heart. By the special connections the SAN tissue is protected from electrical activations arriving from the exterior of the SAN. The general organization of the SAN is said to remain the organization of the embryo heart at the early stage of development [3]. Although the SAN as a whole is recognized as a uniform functional pacemaker unit, only small part of it consists of primary pacemaker cells. This part is called the leading pacemaker site and, for example, in the case of a rabbit it has about 1% of cells [6]. These cells are early generators of the excitation waves which first spread to the rest of the nodal tissue and then are passed to the other parts of the myocardium. Zipes et al. [2] collects the widely accepted results about the electrophysiology of the heart while Mangoni et al. [3] review the results of 40 years of investigations of the SAN.

Despite all complex cellular processes involved in the SAN functioning — the mentioned property of the cell self-excitation and sophisticated ways of signal transduction mechanisms via gap junctions, there are problems related to the arrangement of cells. A cell of the canine SAN has at average $4.8 \pm 0.7$ connections with other cells while any ventricle cell has $11.3 \pm 2.2$ neighbors [8, 9]. It is reasonable to ask how these rather poorly connected cells of the sinoatrial node can drive the heart contraction? It is also known that oscillations produced by the leading pacemaker site are faster than oscillations of an individual cell. Therefore, the intercellular network of interactions should be crucial for the emergence of this phenomena. It is known that the structure of intercellular connections is heterogeneous [9]. Most of the canine SAN cell connections, namely approximately 75% are found to be lateral but a ventrical cell has on average only 37% lateral connections. The lateral connections are known to be less efficient in transmitting electrochemical signals than, e.g., the end-to-end connections.

In the following we propose a model for the SAN tissue. The model considers cells interacting along a network formed from intercellular connections. We propose a construction of a heterogeneous network and then test the role of heterogeneity. Our main objective is to consider whether
the robust SAN automatically is supported by the special topology of intercellular connections. The considerations of this paper continue our earlier research though the present model differs in details from the model presented previously [10–12].

The paper is organized as follows. In Sec. 2 the model of the SAN tissue is presented in two steps: by describing oscillatory dynamics of a cell, and then by designing a network of intercellular connections. In Sec. 3 the phenomenon of persistent entrainment is introduced and its role in producing the shortest oscillations is discussed. Sec. 4 presents results obtained with simulations and Sec. 5 compares our main observations to the biological facts.

2. The model

We refer to [1, 10, 11] for the explanation of how the model is built and how it takes into account physiological facts.

2.1. Modeling of electric activity of cell membrane

A three-state cellular automaton known as the Greenberg–Hastings (GH) cellular automaton is our starting point to model the excitable medium [13, 14]. However we modify the GH model to reconstruct the special properties of the SAN cells. Our proposition follows ideas of [15] and [16] and goes further. Fig. 1 illustrates the dynamics of an individual cell.

Fig. 1. Intrinsic cycle of a cell. Grey (red) arrows indicate at the phase of a cycle when a cell sends signal to other (outward) or reads signals from others (inward).
Formally let $\Sigma^* = \{(\sigma_s) : \sigma \in \Sigma, \ s = 1, 2, \ldots, n_\sigma \}$ be the state space of a cell, where:

- $\Sigma = \{E, R, A\}$ is a discrete state set describing the three main phases of the cellular membrane electrical properties: $E$ — excitation, $R$ — refractory and $A$ — activation phase, subsequently changed in the intrinsic cycle of any self-excited cell: $E \rightarrow R \rightarrow A \rightarrow E \rightarrow \ldots$;

- $next: \Sigma \rightarrow \Sigma$ defines next state in the life cycle of a cell, i.e., $next(E) = R$, $next(R) = A$, $next(A) = E$;

- $s$ counts time steps spent by a cell in a state $\sigma$;

- $n_\sigma$ denotes the maximal time in which a cell can stay in a state $\sigma$; correspondingly, we define $n_E$, $n_R$ and $n_A$, so $n_\sigma \in \{n_E, n_R, n_A\}$;

- at each time step $t$ the value of $n_E, n_R$ and $n_A$ can be shortened with probability, $(s(t)/n_\sigma)^\xi$ where $\xi > 1$ has a fixed value.

$$
\begin{align*}
\left( \begin{array}{c}
\sigma(t+1) \\
s(t+1)
\end{array} \right) &= \begin{cases} 
\left( \frac{next(\sigma(t))}{1} \right) & \text{at rate } (\frac{s(t)}{n_\sigma})^\xi, \\
E & \text{if } \sigma(t) = A \text{ and a cell receives external stimulus not weaker than threshold } T_F, \\
\left( \frac{\sigma(t)}{s(t)+1} \right) & \text{at rate } 1 - (\frac{s(t)}{n_\sigma})^\xi.
\end{cases}
\end{align*}
$$

Hence, in the absence of any external stimulus each cell performs the intrinsic cyclic dynamics. Notice that if $\xi$ is significantly greater than 1 then the intrinsic cycle is rarely shortened and the evolution can be seen as deterministic. In such case the period is

$$
T = n_E + n_R + n_A.
$$

In the case of deterministic dynamics the shortened periods can occur due to interactions with other cells. The shortest possible cycle is

$$
T^* = n_E + n_R + 1.
$$

The intercellular interactions are modeled as follows. A cell becomes excited $E$ in the next time step if at present a cell is in the activation state $A$ and the number of its nearest-neighbors in $E$ state is not smaller than the threshold value $T_F$. 

2.2. Modeling of intercellular connections

The SAN tissue is almost flat and therefore any two-dimensional lattice can be a good starting point. Let \( N \) cells be located in vertices of a square lattice \( N = L^2 \), see Fig. 2. Let interactions between cells take place only via links established between the vertices. The procedure of building a network of intercellular connections goes in two steps. At first, intercellular connections are established at random though with preference set to lateral connections. By this preference the heterogeneity to the network structure is injected, what allows to study the role of domination of lateral connections. Then, in the second step, the connections established are carefully rewired making the lattice structure more similar to the real three dimensional medium. The rewiring algorithm (based on the classical Watts–Strogatz \([18]\) rewiring rule of diffusive type) considers changes between nearest neighbors only. Additionally, while choosing a neighbor to break a connection, the preference is set to neighbors which at the moment have less numbers of neighbors. By this rule, densely connected cells are protected, hence favored. Due to rewiring procedure the heterogeneity is related with density of connections.

Fig. 2. A stochastic network of intercellular connections. The leftmost and rightmost columns are the output cells to other parts of the heart.

To specify formally the above description let us introduce the two following parameters which will control the network topology:

**K:** parameter of stochastic heterogeneity — for a given \( d_{\text{lateral}} \in [0, 1] \) a lateral link on a square lattice is created with a probability \( d_{\text{lateral}} \); a vertical or horizontal link is created with a probability \( d_{\text{lateral}}/K \).

Additionally, cells from the leftmost and rightmost columns are the output cells of the SAN and input cells to the crista terminalis and atria are always connected horizontally to the next cell because horizontal connections are dominant among cells of crista terminalis. Each isolated cell, if happens, is linked to its nearest right neighbor. By these two extra rules some extra horizontal connections appear. Notice that cells from the top
and bottom rows have less connections. This property can be thought of as imitating the connective tissue barrier shielding the SAN from the atria hyperpolarizing influence [5].

Notice that the mean density of edges $d$ on the 2D lattice is

$$d = \frac{d_{\text{lateral}}}{2} \left( 1 + \frac{1}{K} \right),$$

which means that the mean vertex degree is $8d$. Recalling facts known from physiology, mentioned in the Introduction, we see that $d = 0.60$ and $K = 4$ are the values which are referred to the nature.

**J: parameter of intensity of wrinkling** — for a given parameter $p \in [0, 1]$ and a cell $A$, the probability to break a link between cells $A$ and $B$ is $p_{\text{break}} = p / \text{deg}(B)$ where $\text{deg}(B)$ denotes the vertex degree of a $B$ cell. A new cell $B'$ is linked to the cell $A$ in exchange to the cell $B$. $B'$ is chosen at random from the set of actual nearest neighbors of the cell $B$. Breaking the connection with a leaf is forbidden. A random choice of a cell $A$ is repeated $N$ times what is referred as one Monte Carlo (MC) step. The table with information about vertex degrees is updated once in each MC step. In the following we always set $p = 0.01$ to weaken the influence of the limited information about vertex degrees. Eventually, the intensity of rewiring is measured by the number of MC steps $J$ applied to a network.

### 3. Persistent entrainment

Entrainment is the process by which two interacting oscillating systems accept the same period. This mechanism is considered as essential for the achievement of coordination and rhythmic impulse generation by the SAN [17]. In the system we investigate, it is easy to find circumstances when the entrainment takes place. But here there are also possibilities that the process of adjusting oscillatory phases between cells becomes permanent. We will call such cell-to-cell interaction the persistent entrainment.

In the case when the threshold value $T_F$ is equal to 1, the simplest possibility to observe the persistent entrainment is to study a triangle of cells with properly adjusted phases — see Fig. 3 left-top. If the threshold value $T_F$ grows then the more complicated structures are needed to result in the permanent entrainment. In Fig. 3 right, an example of such structure is shown for $T_F = 2$. It consists of 9 cells and it demands locally changed topology.

It is easy to notice that the persistent entrainment needs the presence of cycles (i.e. closed paths in the graph description) in the network of intercellular connections. However, from the physiological point of view the triangular cell-to-cell organizations means wasting of electrochemical energy.
Fig. 3. Left-top: The simplest structure of interactions which can lead to oscillations with $T^*$ on 2D lattice, case $T_F = 1$. Right: Example of a structure which can lead to oscillations with $T^*$ on rewired locally lattice in the case of $T_F = 2$. Black arrows denote regular edges of 2D lattice, gray (green) edges can appear due to rewiring. White (yellow) cells excite gray (green) cells; gray cells excite dark black (blue) cells, and dark blue cells excite yellow cells. Left-bottom: Example of evolution driven by the persistent entrainment between three cells when $n_E = 3$, $n_R = 5$ and any $n_A$.

of a propagated signal. Moreover, since the common period of cells entangled in the persistent entrainment is the shortest one $T^*$, if the evolution is deterministic, then these cells live independently of other neighbors, hence, they block propagations of a signal. So the two SAN aims: being a source of shortest oscillations and efficient propagation of a signal, are opposed each other from the point of view of the network organization. Therefore it is interesting to study whether this conflict can lead to the optimum solution when both aims are robustly and efficiently performed. In particular, we ask how the heterogeneity of intercellular connections influences the optimal solution.

In the following we consider $T_F = 2$ to increase the necessary effort for a system to produce the common effect.

4. Results

4.1. Topological properties of networks

From the construction of the network one can see that, in practice, we deal with networks where heterogeneity is one of three types: (a) because of the dominance of lateral connections: $K > 1$ and $J = 0$, (b) because of rewiring: $K = 1$ and $J > 0$, and (c) initially dominated lateral connections are then rewired: $K > 1$ and $J > 0$. Notice that rewiring leads to emerging cells which are connected to others more densely than it happens on the
ordinary square lattice, namely, their vertex degree is greater than 8. The unwanted effect of the higher intensity of rewirings is that more isolated cells appear, see [10, 11]. In the case of $J = 200$ about 1/3 cells are leaves. Such cells cannot be excited by interactions when $T_F = 2$ because of their isolation. So their presence limits the efficient signal propagation.

Here we concentrate on the cyclic structures. In particular, we investigate the number of triangles on the networks. The results are presented in Fig. 4. It is easy to see that the number of triangles decreases significantly if initial domination of lateral connections grows — bigger $K$ effects, in general, in smaller number of triangles. The intensity of wrinkling $J$ also influences the number of triangles. If all directions are initially almost equivalent then the number of triangles decreases with the increase of $J$ — case $K = 1, 2$. But if initial domination of lateral connections is set — case $K = 3, 4$, then the number of triangles grows though their characteristics stay significantly below the cases $K = 1, 2$. Hence each type of the network (a), (b) and (c) has a different local topology.

![Fig. 4. Distribution of triangles for different heterogeneity parameter $K$ and different intensity of rewiring $J$.](image)

4.2. *Deterministic evolution: $\xi \gg 1$*

Let us assume that the ratio of cells evolving with the shortest period $T^*$ measures the level of the self-organization of oscillatory phases of cells. We simulated systems with the deterministic dynamics of a cell at: $n_E = 10, n_R = 20, n_A = 20$ and $T_F = 2$. At such values $n_E, n_R, n_A$ the dominant frequency is expected to be $T$ (here, $T = 50, T^* = 31$) if a system is considered on a plain lattice [11].
Fig. 5 left shows the distributions of cycle lengths, received from all cells from the networks homogeneous $K = 1$ and not rewired $J = 0$, with respect to the density $d$ of intercellular connections. The distributions were found after 10 000 time steps (to give a system time to stabilize first) during the next 10 000 time steps. We see that when density $d$ grows then the transition between two overwhelming network states is observed. Almost all cells have $T$ period and almost all cells have $T^*$ period. The transition takes place for $0.60 < d < 0.65$ — compare a plot formed by black triangles to a plot formed by hexagons in Fig. 5. The switch between the two periods $T = 50$ and $T^* = 31$ is sharp in the case of output cells, Fig. 5 right. All output cells evolve either with $T$ or with $T^*$.

![Fig. 5. Distributions of cycle lengths of all (left) and output (right) cells in the stationary state. The network is homogeneous $K = 1$ and the rewiring algorithm is not applied $J = 0$. Notice that the vertical axis is a log-type.](image_url)

When the intensity of rewirings grows then the transition moves to the value slightly below $d = 0.60$, see Fig. 6 middle panel. At $d = 0.60$ and $J = 100$ most of output cells follow dynamics with the shortest period. Notice that $J = 100$ means that, at average, each connection is rewired with probability close to 1. Because of this the initial structure of a lattice is significantly modified. If the intensity is too high, e.g., $J = 200$, then other periods appear among output cells (even at $d = 0.75$ 10 % of output cells have $T$ period) which weakens the total effect of the self-organization to beating with the shortest period.

If the system is considered on the heterogeneous network: $K = 4$, but not rewired $J = 0$, then the transition is not observed for available edge densities $d$. But when we apply the wrinkling algorithm then the transition appears at $d$ close to 0.63 even at small intensity of rewirings, namely, if $J = 20$, see
Fig. 6. Distributions of periods in output cells in the case of deterministic evolution. The network is homogeneous $K = 1$ but the intensity of rewiring is increasing 20, 100, 200.

Fig. 7, left panel. Notice that $d = 0.63$ in the case of $K = 4$ means that (according to (2)) all lateral connections are present. When the intensity of rewirings grows then the transition takes place for $0.59 < d < 0.63$. But again if the intensity of rewirings is too high ($J = 200$) then plenty of other periods (unwanted) appear among output cells.

4.3. Stochastic evolution: emergence of leading pacemaker sites

The evolution of real cells cannot be strictly deterministic — it is an obvious remark. Our proposition to randomize the lengths of particular states in the intrinsic cellular cycle seems to be a reasonable solution how to model variability in the period length of each individual cell. It is valuable to start with presenting snapshots of network configurations observed in stationary states for different model parameters, see Fig. 8.
The well-established spiral patterns made of cells being in the same state are claimed to be a sign of a proper propagation of impulses in the cardiac tissue [19]. We see that regular large clusters start to emerge if a network is sufficiently densely connected ($d > 0.60$). Regular spirals are observed at higher densities if $K = 1$. But if a network is heterogeneous because of either preference to lateral connections (case $K = 4$) or due to suitable rewiring intensity $J = 100$ then regular spiral patterns emerge. The sources of the spirals can be considered as the leading pacemaker sites.

4.4. Stochastic evolution — properties of output cells

When investigating details of oscillations in the system driven by stochastic dynamics, it is important to remember that random shortenings of each phase of electric activity of a cell affect the length of a cell cycle. The periods of cells take random values — lower than $T = 50$ or $T^* = 31$. Because of this variability maintaining the permanent entrainment needs special circumstances — set e.g., by topology, and changes in the location of the leading pacemaker site could be present.
In Fig. 9 we present results describing systems evolving on heterogeneous networks which appeared to be best in the case of deterministic dynamics: networks that are initially not heterogeneous $K = 1$ but then become heterogeneous after local preferential rewirings $J = 100$, and networks which initially are dominated by lateral connections $K = 4$ and then wrinkled at $J = 100$. In Fig. 9, together with the distributions of periods found in output cells, we show the distributions of periods in cells that are the most densely connected to the networks — cells for which vertex degree is greater than 8. Such cells are possible sources of the structures allowing for the persistent entrainment.

![Fig. 9. Distribution of periods in output cells and in cell that are densely connected (deg > 8) to the network.](image)

It is easy to read the lengths of dominating cycles for a given densities. In the case of $K = 1$ the transition appears at a density $d$ identical to the one observed in the deterministic case. But if $K = 4$ then the transition to the evolution with the shorter periods appears as the smeared transition —
almost all oscillations are present equivalently among cells of the output and densely connected. The transition takes place if $0.59 < d < 0.63$. One can interpret this result that the transition means here admitting an evolution with a wide spectrum of possible oscillations. It is possible only if different lengths of phase $A$ are permanently present in the system. Such property is extremely important for the autonomic regulation of the heart rate [1, 2].

5. Conclusions

From biological investigations it is known that the SAN structure is different from the rest of the heart. There is no controversy among researchers about rare density of cells and heterogeneity of the SAN tissue. In the presented article we proposed a bio-inspired model of the SAN which allowed to investigate whether and how topology of intercellular connections could influence the functionality of the SAN. Specially, we concentrated on two problems: how density of intercellular connections affected the activity of the SAN, and what was the role of heterogeneity in the intercellular connections. If the heterogeneity was modeled in two ways: by preferences set to the lateral connections or/and by local preferential rearrangement of connections.

By using the cellular automata approach we always strongly oversimplify modeled phenomena. However, we believe that we were still able to learn valuable facts about the modeled system [20]. In simulations we found that the density $d = 0.60$ of intercellular connections was critical for the properties of the modeled phenomena. This critical value corresponds very well to the known value of cell-to-cell connections 4.8 in a dog as it was described in the Introduction. However large and usually individual spirals emerged robustly only if the network of connections was heterogeneous in a special way — the reason for the heterogeneity was the connection’s rewiring. The local rewiring spreads the influence of an individual cell to other cells than a square lattice nearest neighbors. Such connections have not been found in the mentioned biological observations what may suggest the our modeling contradicts to solutions found by the nature. But the variability among the SAN cell shapes (namely, spindle, elongated, spider [3]) is so large when compared to other myocytes, then the standard classification of types of intercellular connections considered by Luke et al. [8] can be confusing.

Many thanks to G. Graff, J. Kaczmarzyk, A. Kolesiak, A. Posiewnik, and M. Żarczyńska-Buchowiecka for fruitful discussions. Authors acknowledge financial support of the Rector of Gdansk University project: BW5400-5-0169-9.
REFERENCES


