

Study of the Preautomatic Pause under Exposure to Acetylcholine in True Pacemaker Cells of Rabbit Sinus Node Using Computer Simulation

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Preautomatic pause, which is required for the restoration of automatism in pacemakers, plays a key role in heart functioning. In this work, we studied the effect of acetylcholine and the role of intracellular ion homeostasis on the occurrence of preautomatic pause in true pacemaker cells of rabbit sinus node. It is demonstrated that, in the absence of acetylcholine, the pause is only 0.4 s, whereas in the presence of acetylcholine it may last for tens seconds. The occurrence of the pause and escape from it is determined by slow changes in intracellular concentrations of Na^+ , K^+ , and Ca^{2+} . Underthreshold fluctuations in membrane potential of increasing amplitude are the sign of automatism restoration.

Conditions of numerical experiments. The electrical activity of membranes of sinus-node cells and the effect of acetylcholine were studied in the model based on rabbit SN cells [1, 2]. When simulating the intracellular ion homeostasis, we took into account the changes in the concentration of Na^+ , K^+ , and Ca^{2+} in the cell and the function of sarcoplasmic reticulum (SR) [3, 4]. Electrical stimulation of cells was performed periodically ($T = 200$ ms) with 10-ms pulses at a current strength of 80 pA. The total duration of measurements was 265 s, including 5 s of control, 60 s of electric stimulation, and 200 s of observation of preautomatic pause development.

Results. A long-term high-frequency electric stimulation of the sinus node resulted in the development of a preautomatic pause, whose duration depended on the presence of acetylcholine and the intracellular ion homeostasis (Fig. 1). In the absence of acetylcholine or under model conditions at fixed intracellular concentrations of Na^+ , K^+ , and Ca^{2+} , the duration of preautomatic pause was approximately 120% of the period of a normal rhythm. In the presence of acetylcholine, the duration of the pause was tens of seconds. As can be seen in

Fig. 1, 20 nM acetylcholine caused the development of such a pause. Long-term measurements of the potential and intracellular concentrations of Na^+ , K^+ , and Ca^{2+} (Fig. 2) show that the concentrations of Na^+ and K^+ gradually increased and decreased, respectively, upon stimulation, whereas the concentration of Ca^{2+} was rapidly adjusted to new steady-state values and practically did not change until the end of stimulation (Fig. 2, $t = 5$ –65 s). After the cessation of stimulation, the direction of changes in the concentration of Na^+ and K^+ altered to the opposite, and the fluctuations in the transmembrane potential and intracellular Ca^{2+} were not observed ($t = 65$ –132 s). Note that the steady-state transmembrane potential (approximately -40 mV) is close to the inactivation potential of L-type calcium channels, which prevents the activation of sinus-node cells [2]. A slow drift in the concentrations of Na^+ and K^+ resulted first in underthreshold and then in full-amplitude fluctuations in the transmembrane potential and Ca^{2+} concentration (Fig. 2, $t = 132$ –250 s). It should be noted that, after the restoration of automatism, the concentrations of Na^+ and K^+ changed the direction of drift once again, slowly approximating their steady-state values [4]. The process of escape from the preautomatic pause is shown in more detail in Fig. 3. This figure clearly illustrates the underthreshold fluctuations of the transmembrane potential and Ca^{2+} preceding the restoration of automatism, as well as the slight changes in the concentrations of Na^+ and K^+ during each cycle of fluctuations.

Discussion. The preautomatic pause lasting for tens seconds is usually observed in pacemakers of the second and third order. For example, in the case of suppression of automatism of ventricular pacemakers, the pause lasted for nearly 60 s [5]. That long pause in mammalian sinus node is evidently a dangerous pathology. Computed simulation showed that, in the absence of acetylcholine, the preautomatic pause is not observed. However, there are grounds to assume that, under natural conditions of stimulation of sinus-node cells through the vagus nerve, acetylcholine concentrations comparable to those used in this study may appear in the immediate vicinity of sinus-node cells. In this case, preautomatic pause may occur under natural con-

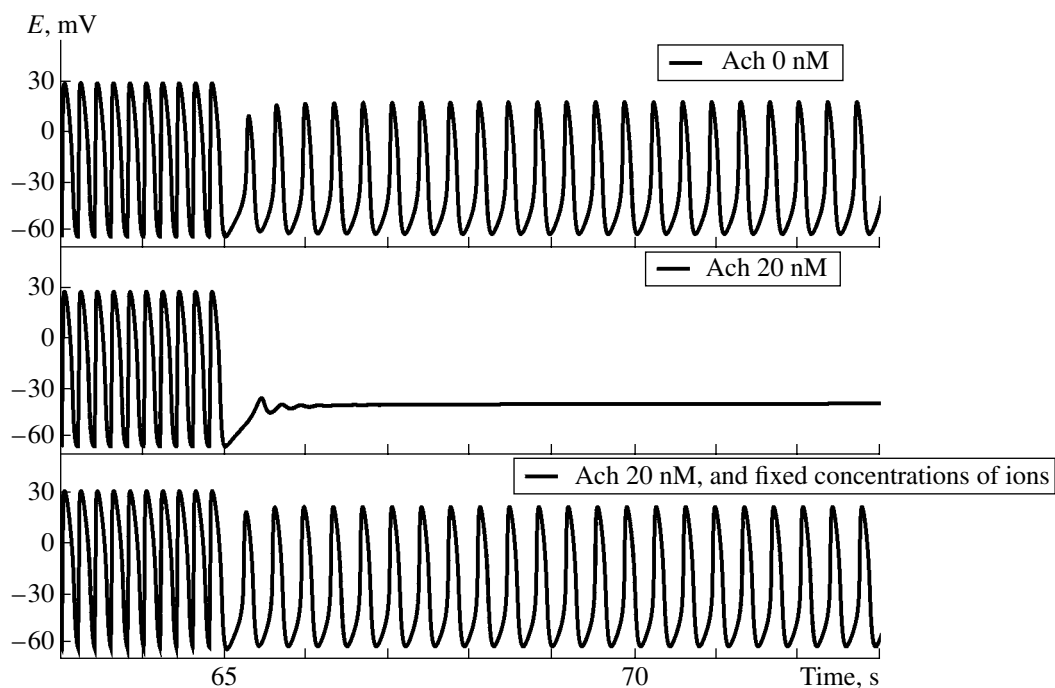


Fig. 1. Preautomatic pause in the absence of acetylcholine, in the presence of 20 nM acetylcholine, and at fixed intracellular concentrations of Na^+ , K^+ , and Ca^{2+} . It is seen that a long-term pause is only developed in the presence of acetylcholine and as a result of change in the concentrations of intracellular ions.

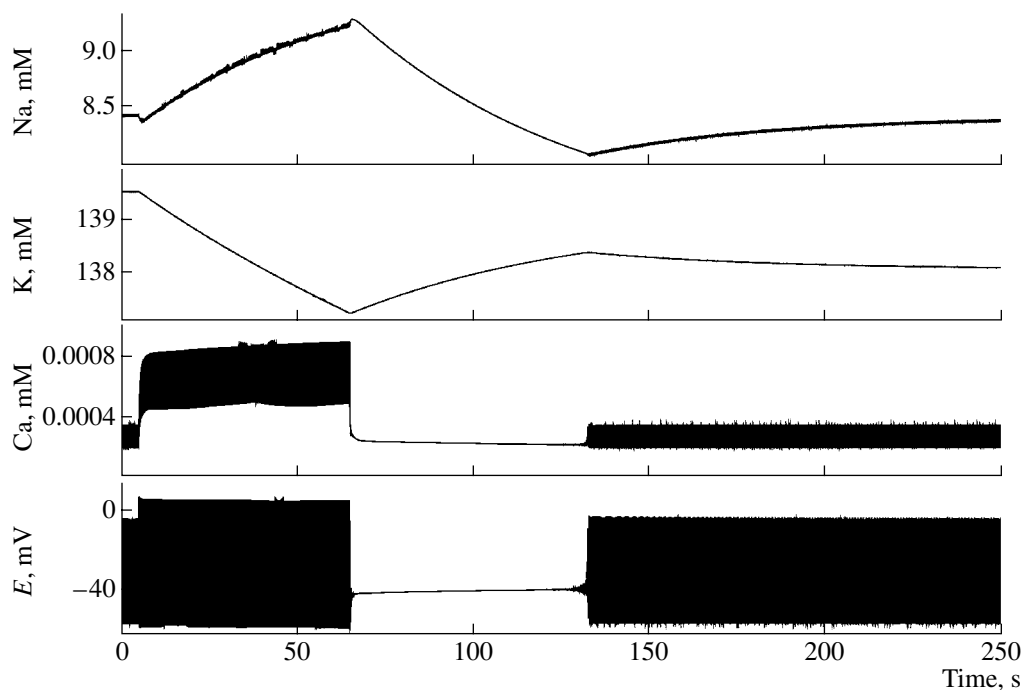


Fig. 2. Intracellular concentrations of ions and the transmembrane potential during development of the preautomatic pause and after restoration of automatism.

ditions of heart functioning as well. The duration of the pause is largely determined by the concentration of acetylcholine. The suppressing effect of acetylcholine on the sinus node is well known: at small concentrations,

acetylcholine decelerates the rhythm, and at greater concentrations it causes termination of spontaneous activity [1, 2]. In this study, we showed that the mechanism of preautomatic pause consists in transient termi-

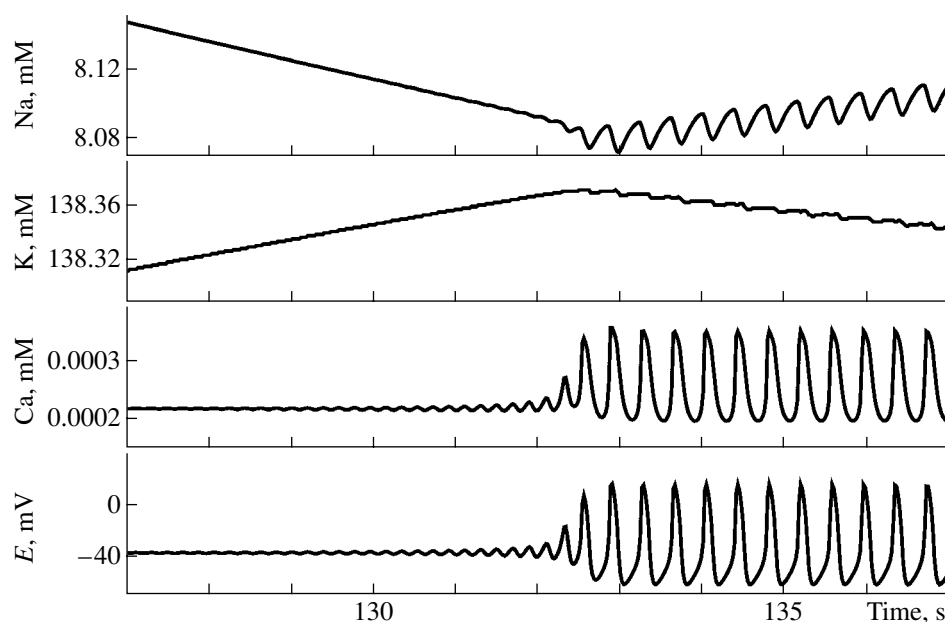


Fig. 3. Fluctuations in the concentration of ions and transmembrane potential in the case of restoration of automatism after the preautomatic pause.

nation of spontaneous activity in sinus node cells in the presence of acetylcholine. Cells escape from the arrest due to slow changes in the intracellular concentrations of Na^+ and K^+ , similarly to the processes of homeostasis onset described in our previous article [4]. It should be also taken into account that these results were obtained using a single cell, and the duration of the pause was maximal. Indeed, the a group of sinus-node cells inhomogeneous either with respect to their properties or environmental conditions, the duration of the preautomatic pause will be determined by the minimal pause among all cells.

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REFERENCES

1. Aliev, R.R., Fedorov, V.V., and Rozenshtaukh, L.V., *Dokl. Akad. Nauk*, 2004, vol. 397, no. 5, pp. 697–700 [*Dokl. Biol. Sci. (Engl. Transl.)*, vol. 397, no. 5, pp. 288–291].
2. Aliev, R.R., Fedorov, V.V., and Rozenshtaukh, L.V., *Dokl. Akad. Nauk*, 2005, vol. 402, no. 4, pp. 223–225.
3. Kurata, Y., Hisatome, I., Imanishi, S., and Shibamoto, T., *Am. J. Physiol.*, 2002, vol. 283, pp. H2074–H2101.
4. Aliev, R.R. and Chailakhyan, L.M., *Dokl. Akad. Nauk*, 2005, vol. 402, no. 5, pp. 236–239.
5. Babskii, E.B. and Saidkarimov, S.K., *Dokl. Akad. Nauk SSSR*, 1970, vol. 191, no. 6, pp. 1420–1423.