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Morphofunctional Organization of Sinoatrial Node in Rat Heart

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The distribution of pacemaker cells along the sinus node artery was studied under conditions of short-term culturing using intracellular glass microelectrodes. The functional borders of the central and peripheral parts of the sinoatrial node were determined. The relationship between the position of the central part of the sinoatrial node and the patterns of the sinus node artery branching were analyzed.

Key Words: *sinoatrial node; pacemaker cells; sinus node artery; intracellular lead; action potentials*

The sinoatrial node (SAN) in mammalians, including humans, is located close to the artery, which is called the sinus node artery (SNA). SAN in rat heart is located around and along SNA. The sources of blood supply and location of the artery are studied in detail, the relationship between specialized nodal cells with SNA *t. media* smooth myocytes is disclosed. Morphofunctional identification using intracellular lanthane label, carried out previously, showed that typical nodal cells corresponded to the true pacemaker cells (PMC) [3]. The SAN periphery is occupied by latent PMC. Linear disposition of SAN along SNA allowed unidimensional mapping of the central part of SAN [1,2].

We studied functional borders of rat SAN and their relationship with the anatomical features of SNA location.

MATERIALS AND METHODS

Experiments were carried out on male Wistar rats (60-90 g). The chest was opened under Nembutal narcosis (40 mg/kg) and 10 ml 0.01% Trypan Blue in Hanks solution (pH 7.35, 37-38°C) was slowly injected into the left ventricle, after which the heart was removed and put into a cuvette with the same solution (15-20°C), the right atrial region containing the anterior wall, right cranial and caudal venae cavae, and the auricle was isolated. SAN was located at the interface between vena cava superior and auricle near SNA, its location was shown by Trypan Blue staining.

The preparation was fixed and placed in a thermocontrolled flow chamber filled with modified Krebs—Ringer solution equilibrated with 5% carbogen to pH 7.4 at 38°C. The rate of medium replacement in the cuvette was 1.7 ml/min. The shape of action potentials of the true and latent PMC was determined using glass microelectrodes. The position of the microelectrode tip was determined with a ruler scale inserted in the microscope objective (1 point on the ruler corresponded to 0.0235 mm).

The shape of PMC action potential was the criterion of true PMC (Fig. 1, *a*): slow diastolic depolarization phase (phase 4), smooth transfer from slow diastolic depolarization phase into initial rapid elevation of potential (phase 0), and slow velocity of the initial rapid elevation of the potential [1,2,4]. Latent PMC are also characterized by diastolic elevation, but differed by abrupt transition from phase 4 into phase 0 and higher rate of potential elevation in phase 0 [4].

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The distribution of different types of PMC action potentials was mapped by consecutive insertion of microelectrodes along SNA (Fig. 1, *b*). The location of true PMC (position of the dominant pacemaker region) was taken for the zero reference point. Positive measuring was carried out down the SNA (similarly with the bloodflow course), negative measuring was made upwards. The extreme points at which the diastolic elevation was still detectable were considered as the lower and upper SAN borders.

The results were statistically processed using Student's *t* test.

RESULTS

Vital staining with Trypan Blue showed that SNA was located at the interface between the right cranial vein and right auricle, was directed towards the caudal vena cava, to the site of SNA thinning and branching small arterioles. Individual features of artery branching can be classified into 4 variants (Fig. 2). The absence of visually seen branching or caudal branching (6 cases) did not indicate the location of the dominant pacemaker region, which could be detected only electrophysiologically (Fig. 2, *a*, *b*). By contrast, cranial branching of the artery (Fig. 2, *c*) coincided with electrophysiologically detected location of the dominant pacemaker region (9 cases). In 2 cases the dominant pacemaker region was situated above the dichotomy, but no more than 0.2 mm higher (Fig. 2, *d*).

Previous studies of dominant pacemaker region migration in response to norepinephrine [1] and acetylcholine [2] showed functional borders of the central part of SAN. The presence of the functional core (nor-

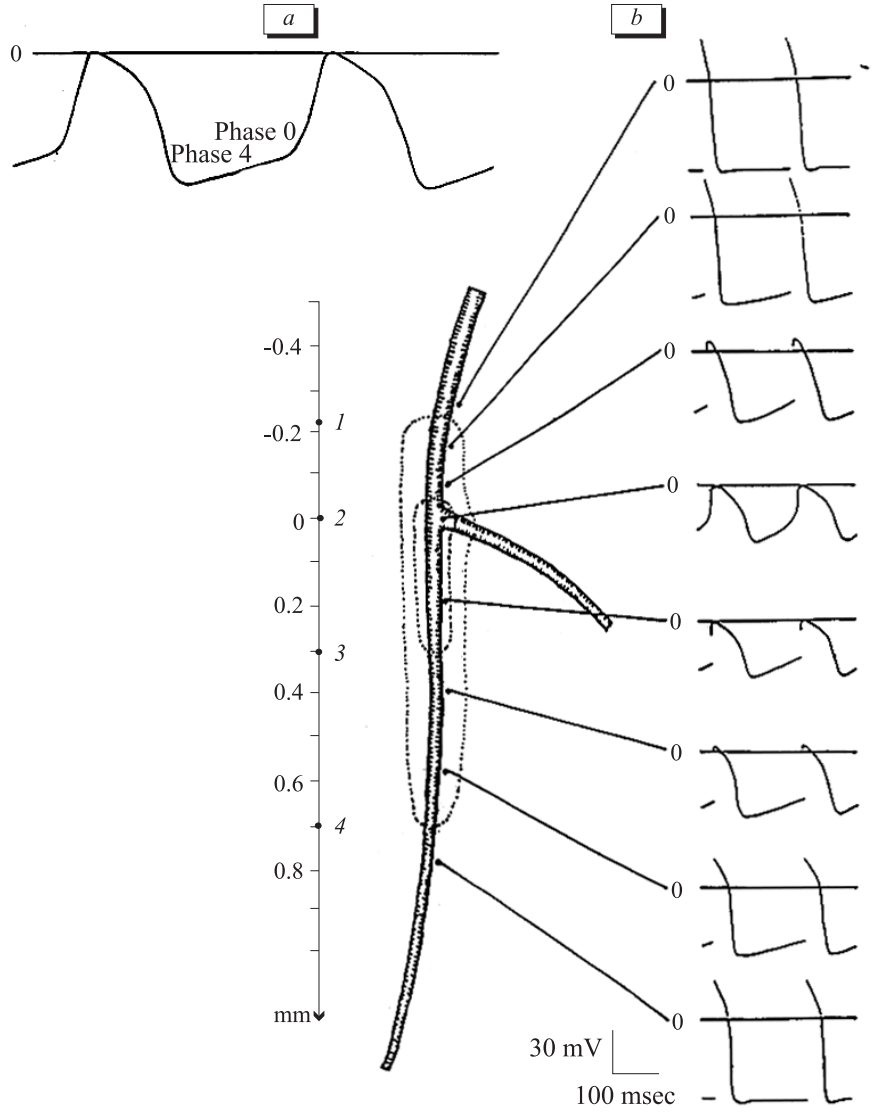

Fig. 1. Action potential of the true pacemaker cell (*a*) and a variant (*b*) of distribution of electrophysiological types of cells along the sinus node artery (type of artery branching is shown in Fig. 2, *c*). *1*) upper border of the sinoatrial node (SAN) (-0.23+0.05 mm; $p<0.05$; $n=17$); *2*) location of the true pacemaker cells; *3*) lower border of SAN central part (about 0.3 mm); *4*) lower border of SAN (0.70+0.10 mm; $p<0.05$; $n=17$).

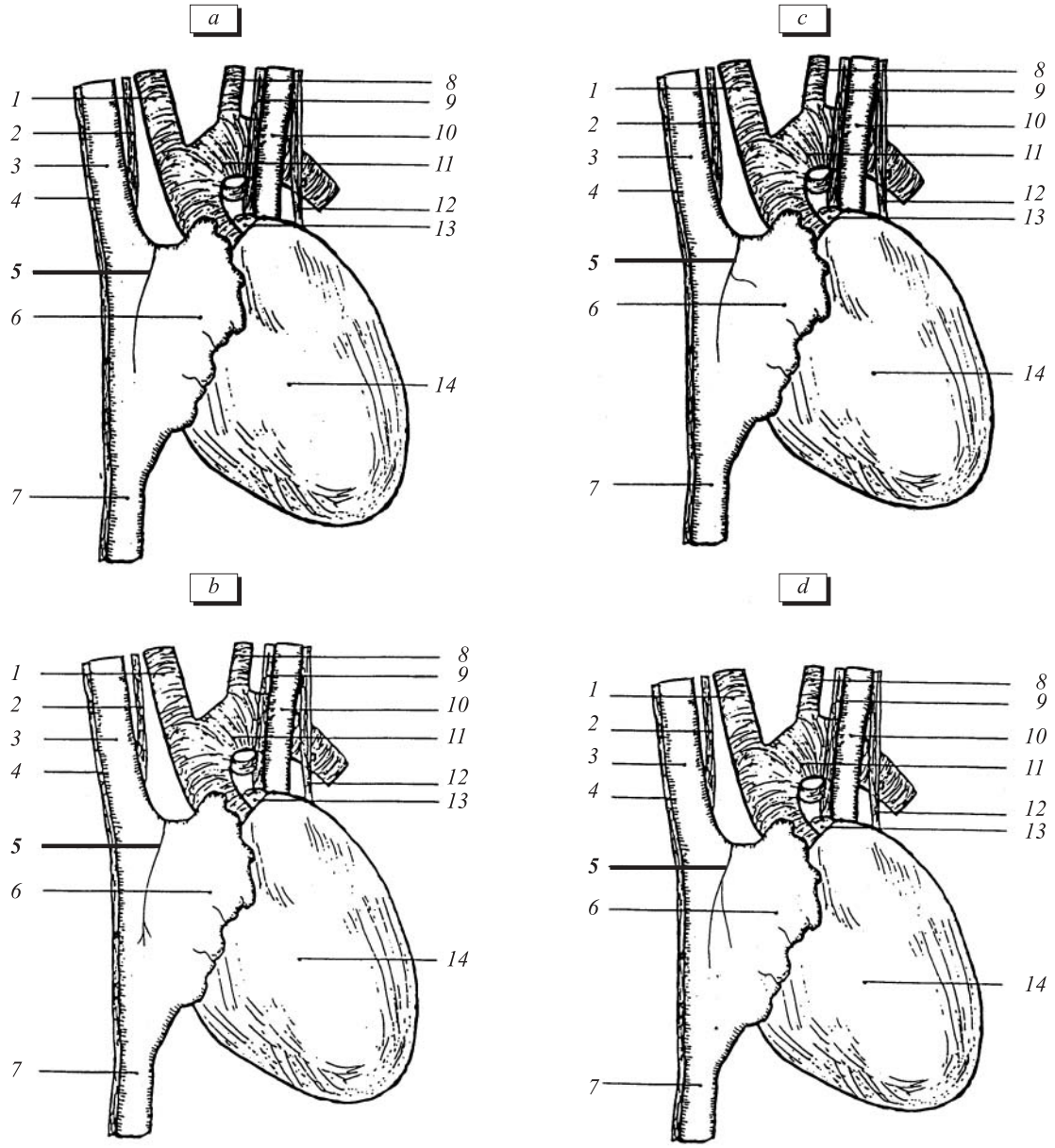

Fig. 2. Variants ofsinus node artery branching in rat heart. *a*) no visible branching (*n*=4); *b*) caudal branching: the artery is divided into several small branches at once (*n*=2); *c*) cranial branching of the artery, a stem of a lesser diameter going virtually perpendicularly towards the right auricle (*n*=5); *d*) cranial branching of the artery, the artery is divided into 2 more or less equal vessels at a small angle (*n*=6). *1*) *truncus brachiocephalicus*; *2*) *n. vagus dexter*; *3*) *v. cava cranialis dextra*; *4*) *n. phrenicus dexter*; *5*) sinus node artery; *6*) *auricula dextra*; *7*) *v. cava caudalis*; *8*) *a. carotis communis sinistra*; *9*) *n. vagus sinister*; *10*) *v. cava cranialis sinistra*; *11*) *arcus aortae*; *12*) *n. phrenicus sinister*; *13*) auricular sinistra; *14*) *ventriculus dexter*.

mal position of the region) and functional tail, within which the dominant pacemaker region could migrate, were demonstrated. The core and tail together (central part of SAN) occupy about 0.3 mm along SNA.

The regularities of changes in the shape of action potential with increasing the distance from the central part of SAN were similar: gradual increase in action potential amplitude, potential reversion, more sharp increase of the potential during phase 0, more smooth diastolic depolarization slope (potential increase during phase 4), and narrowing of the peak of the action potential. Similar changes in the shape of action potentials were observed in other mammal [4]. The functional border of the SAN peripheral part in the downward direction is several times greater than the upper border, that is, the peripheral part of SAN in fact repeats the shape of the central part, including and encircling it. Since the peripheral part of SAN seems to

be not involved in the nervous regulation of cardiac chronotropy (all effects, including transposition of the dominant pacemaker region, were realized in the central part [2]), presumably, its main function is protection of the central part of SAN from electrotonic effect of atrial myocytes [4].

The relationship between the dominant pacemaker region and the site of SNA branching is worthy to note. In 9 of 11 cases with cranial branching of SNA this region was situated precisely at the site of dichotomy, in 2 cases it was above the site of branching. It means that the site of dichotomy is by all means within the central part of SAN (within the functional tail). Taking into account the high incidence of cranial branching of SNA (in our experiment in 11 of 17 cases), it can serve as the morphological marker of position of the central part of rat heart SAN. The cause of this phenomenon can become the object of further studies.

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