EFFECT OF COOLING ON RHYTHM-CONTRACTION RELATIONS IN THE PATHOLOGICALLY CHANGED MYOCARDIUM

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The introduction of cardioplegia on a wide scale into cardiac surgery has necessitated a close study of the effect of the different components of the cardioplegic solution and the temperature at which the heart is kept on myocardial contractile function after the end of the cardioplegic pause. Direct proof of changes in contractility of the heart after cardioplegia is extremely difficult to obtain on the intact organism, and information on this matter in the literature is based on indirect estimations [5, 11]. It has been shown that frequency–strength relations in the heart can serve as an indicator of the contractile function of the human myocardium [1, 7]. Accordingly, to obtain direct information on the effect of solution temperature on the myocardium under continuous oxygenation conditions, frequency–stregth relations were investigated in fragments of myocardium from patients undergoing operations for congenital and acquired heart disease, before and after the hypothermic pause.

The object of this investigation was to study the effect of the temperature of the oxygenated hypothermic solution on recovery of contractility of the myocardium during its reheating, and also the effect of cardioactive drugs (dopamine, isoproterenol, strophanthin) on the myocardium before and after the hypothermic pause.

EXPERIMENTAL METHOD

Experiments were carried out on fragments of the auricle of the right atrium removed before connection to the artificial circulation apparatus from 40 patients, of whom 23 were undergoing operations for rheumatic heart disease and 17 for congenital defects.

The removed auricle was placed in oxygenated Tyrode solution and sent to the laboratory in a container. After transportation for 15-20 min the auricle was transferred to a dissection bath containing Tyrode solution, enriched with carbogen (95% O_2 + 5% CO_2). Trabeculae 3-5 mm long, 1-2 mm wide, and not more than 1 mm thick were removed from the auricle. Transportation and dissection of the trabeculae were carried out at room temperature. The isolated trabeculae were placed in a working chamber perfused with Tyrode solution, continuously saturated with carbogen (32-34°C). One end of the myocardial preparations was fixed securely to a metal hook built into the floor of the chamber, and the other end was fixed by means of a ligature to the rod of a 6MKhlS mechanotron. The output signal from the mechanotron was led to the input of a dc amplifier, from which it was led to a recorder (S1-18 oscilloscope). In the original state the myocardial preparation had no spontaneous activity, and for that reason the trabeculae were stimulated by means of silver disk electrodes. Square pulses of above threshold strength and 10-100 msec in duration were applied. At the beginning of each experiment the preparation was stimulated with a frequency of 0.5 Hz for 1-1.5 h. Rhythmic contractions were then recorded as the frequency of stimulation was progressively changed from 0.1 to 2 Hz. Stimulation of the preparation was then stopped for 60 min (hypothermic pause) and the resting potential was recorded by means of a KSP-4 automatic writer. During the pause perfusion of the preparation continued with oxygenated Tyrode solution. The temperature of the solution in the chamber during the pause was regulated by means of a microrefrigerator. In the experiments of series I the temperature of the solution during the pause

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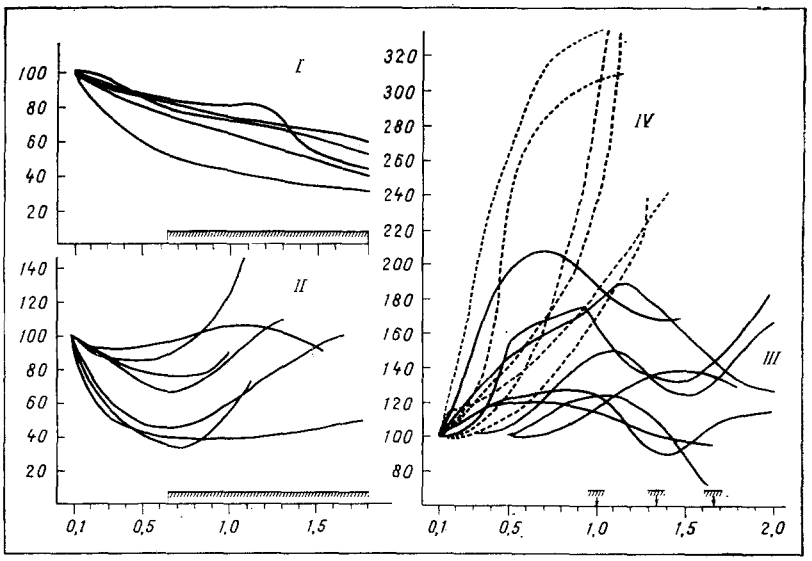

Fig. 1. Types of frequency—strength relations in the patients' myocardium. Here and in Figs. 2 and 3: abscissa, frequency of stimulation of preparations (in Hz); ordinate, amplitude of contractions (in %). Amplitude of contractions with frequency of stimulation of 0.1 Hz taken as 100%. Shaded strip along abscissa indicates range of frequencies determined from ECG in heart of patients whose myocardium was being studied.

was unchanged (32-34°C), and in series II and III it was lowered during the pause to 24-26 and 10-14°C respectively. Since the diameter of the myocardial trabeculae was 1 mm, the temperature of the solution and of the myocardium was the same. After the end of the pause the temperature of the Tyrode solution was restored to its initial value and stimulation of the preparation resumed with a frequency of 0.5 Hz. After 30 min control measurements of the rhythmic contractions were made. The Tyrode solution had the following composition (in mM): NaCl — 131, KCl — 4.5, $NaHCO_3$ — 11, NaH_2PO_4 — 0.6, $MgCL_2$ — 0.25, $CaCl_2$ — 2.16, glucose 11.0, pH 7.4. The action of dopamine (10^{-6} g/ml), isoproterenol (10^{-7} g/ml), and strophanthin (10^{-6} g/ml) was investigated before and after the hypothermic pause.

EXPERIMENTAL RESULTS

Four types of frequency—strength curve characterizing these relationships in the patients' myocardium are illustrated in Fig. 1. Curves of types I, II, and III were found in the myocardium of patients with rheumatic heart disease. In some cases, if a severe form of Fallot's tetralogy was present, the patients also gave a type III curve. The type IV curve was found only in the myocardium of patients with congenital heart disease, with no visible signs of changes in the myocardium. The shape of the curves of types I, II, and III, as the writers showed previously [2], is connected with the development of poststimulation contractile oscillations, modifying the amplitude of contractions evoked by electrical stimulation.

A pause of 60 min in Tyrode's solution of the original temperature caused no changes in the resting tension of the myocardial preparations. Cooling the myocardium during the pause to 24-26°C led to slow development of a contracture, which was increasingly more marked as the temperature of the myocardium fell to 10-14°C. Rapid heating of the preparation to the original temperature led to the appearance of an additional peak of mechanical tension with a rapid rising phase and slow falling phase. In four preparations of myocardium with type II frequency—strength curve, spontaneous activity developed during the pause when the myocardial temperature was 24-26°C. To abolish it, the potassium ion concentration in the solution was increased gradually, starting from an initial concentration of 4.5 mM. A K^+ concentration of 7 and 9 mM did not abolish the spontaneous activity. The minimal K^+ concentration which inhibited spontaneous activity of the preparations was 13.5 mM. The development of contracture in the period of the hypothermic pause was independent of the type of frequency—strength relations in the myocardium.

TABLE 1. Changes in Amplitude of Rhythmic Contractions (in %) of Fragments of Human Myocardium after Interruption of Stimulation for 60 min in Hypothermic Solution at Different Temperatures (M ± m)

Type of disease	Period of investigation	Period of investigation		Frequency of stimulation, Hz						
				0,1	0,5	0,7	1,0	1,2	1,6	2,0
Congenital	32—34	Initial value	7	100,0	83,1±5,1	92,2±6,9	84,4±6,9	—	—	123,0±14,2
		After pause	6	104,0±31,3	96,0±32,1	74,4±31,5	61,8±22,5	—	—	79,0±21,0
Acquired	32—34	Initial value	6	100,0	78,2±5,1	77,0±6,6	73,5±8,2	68,5±9,5	49,0±6,9	—
		After pause	6	67,6±9,6*	66,0±9,7	63,2±9,1	67,3±9,4	63,8±9,0	48,5±7,7	—
Acquired	24—26	Initial value	4	100,0	67,7±14,6	74,2±19,6	90,2±25,6	88,3±32,2	87,3±22,2	—
		After pause	(4)	103,2±8,6	63,5±15,9	60,2±16,9	61,7±16,5	79,5±30,6	91,7±42,0	—
Congenital	10—14	Initial value	9	100,0	99,5±20,4	93,0±10,0	99,6±13,6	—	—	124,6±12,3
		After pause	7	62,4±16,1*	44,4±13,7*	44,4±11,7*	39,8±10,5*	—	—	27,3±19,4*
Congenital	12—14	Initial value	7	100,0	101,6±3,1	105,4±4,1	109,6±5,8	121,7±10,4	—	118,0±12,0
	K^+—22 mM	After pause	7	101,5±14,7	82,6±10,3*	72,3±9,1*	86,3±12,6*	90,4±16,9	—	122,0±46,0
Acquired	12—14	Initial value	7	100,0	72,6±6,4	61,6±5,3	59,0±7,2	59,6±4,4	—	—
	K^+—22 mM	After pause	7	49,7±5,7*	42,3±7,1*	39,7±5,9*	35,3±5,9*	43,0±9,2	—	—

Legend. *P < 0.05; elsewhere differences are not significant (P > 0.05).

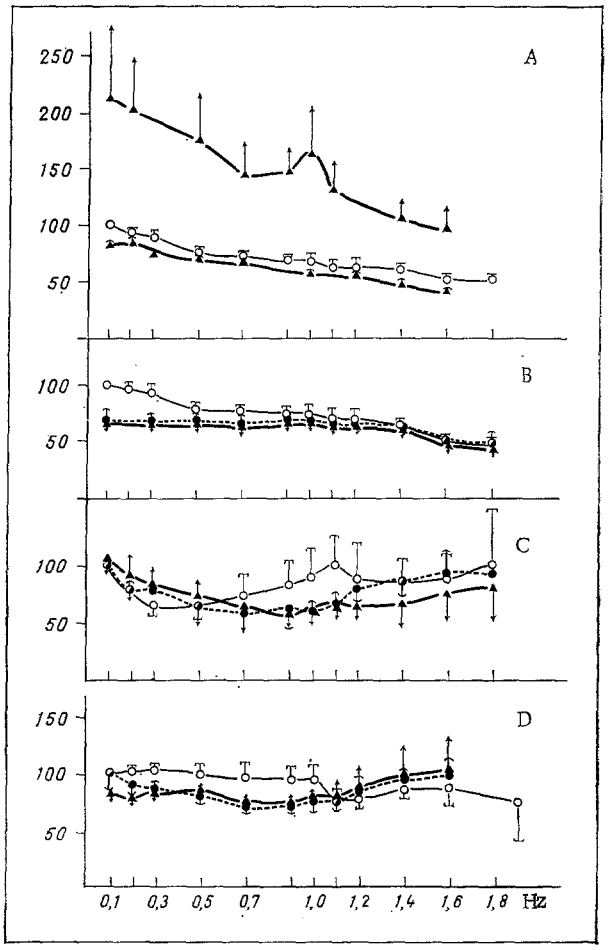

Fig. 2

Fig. 2. Effect of dopamine on frequency—strength relations in myocardium of patients with acquired heart disease. A) Reaction to dopamine before pause (top curve — two preparations tested, bottom curve — four preparations); B-D) hypothermic pause of 60 min at temperature of 32-34°C (six preparations), 24-26°C (four), and 12-14°C (five) respectively. Empty circles denote original frequency—strength relations; filled circles — after pause of 60 min; triangles — after dopamine (10^{-6} g/ml).

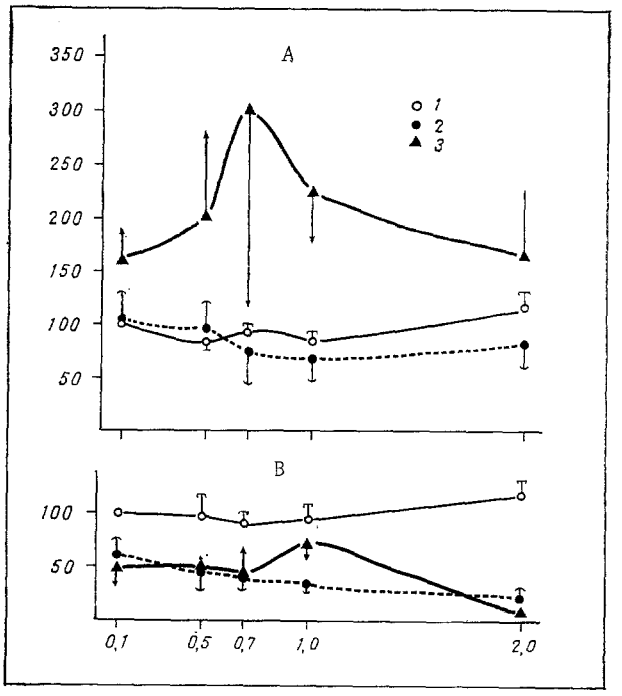

Fig. 3

Fig. 3. Effect of strophantin on frequency—strength relations in myocardium of patients with acquired diseases. Temperature of hypothermic pause: A) 32-34°C; B) 10-14°C. Empty circles — original frequency—strength relations; filled circles — after pause of 60 min; triangles — strophanthin (10^{-6} g/ml).

Data on the original steady-state amplitude of rhythmic contractions of the myocardial preparations, expressed as percentages of its changes after a 60-min pause, are given in Table 1. The original amplitude of contractions during stimulation at a frequency of 0.1 Hz was taken as 100%. As Table 1 shows, if the temperature of the solution remained unchanged during the pause (32-34°C), no significant changes in the amplitude of the rhythmic contractions took place in preparations taken from patients with both congenital and acquired heart diseases ($P > 0.05$). No significant changes in contractile activity of the myocardium likewise were found after the temperature of the solution had been lowered to 24-26°C. The amplitude of contractions was significantly reduced at all frequencies of stimulation of the preparations after the pause if the temperature of the solution was 10-14°C. A fivefold increase in the potassium ion concentration (up to 22.5 mM) in the solution during the pause at a temperature of 12-14°C partially abolished the inhibitory action of cooling for 50% of the preparations. No significant decrease in the amplitude of contractions took place in these experiments at frequencies of stimulation above 1 Hz (Table 1). This effect was found

during the study of myocardial preparations from patients of both groups. However, in the remaining experiments, with an excess of potassium ions in the solution, the inhibitory action of cooling was preserved.

Before the hypothermic pause, dopamine, isoproterenol, and strophanthin increased the amplitude of contractions of the myocardial trabeculae from patients with congenital heart diseases; after the pause, the positive inotropic action of the cardioactive drugs still remained in this series of experiments. In preparations taken from patients with acquired diseases (Fig. 2A), dopamine caused the development of a positive inotropic effect (top curve) in only two of six cases, and in the other four (bottom curve) the amplitude of contractions was actually reduced a little after addition of dopamine compared with the initial value (middle curve). Dopamine did not exhibit a positive inotropic action after the 60-min pause (Fig. 2B-D), irrespective of the temperature of the solution. Isoproterenol had a positive inotropic effect before the pause, but after the pause an increase in amplitude of the contractions was observed in only three of seven preparations. Strophanthin, however, did not lose its positive inotropic action on the myocardium after a pause at a temperature of 32-34°C. If, however, the temperature during the pause was lowered to 10-14°C, although strophanthin increased the amplitude of contractions (at a frequency of stimulation of 1 Hz), these contractions did not reach the original level (Fig. 3).

Four types of frequency—strength relationship could thus be conventionally distinguished in the myocardium of patients with congenital and acquired heart diseases. Rhythmic contractions in both types of diseases were found not to undergo any statistically significant changes after interruption of stimulation for 60 min at temperatures of between 34 and 24°C. Lowering the temperature of the myocardium during the pause to 10-14°C caused the development of a contracture and led to considerable inhibition of contractions after reheating of the preparations and resumption of their stimulation. The main cause of development of the contracture and of inhibition of the amplitude of contractions of the myocardial preparations during cooling was probably inhibition of active transport of Ca^{++} ions (sequestration) in the cardiac cells. The results agree with those obtained by other workers [6], who found that recovery of the contractile function of the left ventricle in cats after a 60-min cardioplegic pause takes place better if the temperature of the myocardium is 27°C.

It was shown previously that an excess of K^+ in the hypothermic solution partially protects the myocardium against the inhibitory action of cold [10]. Very probably this phenomenon is connected with prevention of loss of intracellular K^+ due to inhibition of Na,K-ATPase during cooling.

Absence of a positive inotropic action of dopamine on the myocardium of most patients with acquired heart diseases, revealed by these experiments, was due to the low sensitivity of the myocardium of patients with rheumatic heart diseases to catecholamines [4] and was probably connected with the lowering of β-reactivity in hypertrophy of the myocardium [12]. The cause of the absence of an effect of catecholamines after the long pause has not yet been explained. It may be due either to desensitization of α- and β-adrenoreceptors or to a disturbance of electromechanical coupling. Weakening of the response to strophanthin after cooling of the myocardium for 60 min to 10-14°C evidently takes place through a decrease in activity of the enzyme systems [8, 9] concerned in the mechanism of action of cardiac glycosides on the heart [3].

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EFFECT OF CEREBELLECTOMY ON A STRYCHNINE FOCUS IN THE CEREBRAL CORTEX

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In order to inhibit epileptic activity and also to cut short other neuropathological syndromes characterized by hyperactivity of pathological systems [1], a factor of great importance is activation of physiological antisystems, which are the functional opposite of structures involved in the pathological process [2, 3]. A component of the antiepileptic system is the cerebellum, activation of which can lead to the suppression of epileptic discharges in the cerebral cortex [6]. However, data on the effect of electrical stimulation of the cerebellum on epileptogenesis are ambiguous [4, 7, 8].

The aim of the investigation described below was accordingly to study the effect of cerebellectomy on the duration of existence of an epileptogenic focus, which reflects in a general manner the character of participation of the cerebellum in the epileptization of the brain.

EXPERIMENTAL METHOD

Acute experiments were carried out on 12 cats weighing 2.5-3.5 kg. Under ether anesthesia tracheotomy was performed, and the skin and subcutaneous cellular tissue was divided by a midline incision from the nasal bones to the occiput. The cranial bones were trephined in the frontal and occipital regions to provide access to the sensomotor areas of the neocortex and the ipsilateral surface of the cerebellum. After the dura had been opened an epileptic focus was created in the posterior sigmoid gyrus by application of a piece of filter paper (2 mm^2) soaked in 0.1% strychnine nitrate solution. The piece of paper with strychnine was removed in all cases 1 min after development of seizure potentials. The life of the foci was measured from the time of appearance of the first spike to disappearance of the last spike. After two or three such determinations the cerebellum was aspirated under visual control. Another strychnine focus was then created in the same way 40-60 min after aspiration of the cerebellum.

Cortical potentials were recorded by a monopolar method on the 4-ÉÉG-3 ink-writing electroencephalograph, the reference electrode being fixed in the nasal bones. The results described in this paper were obtained in experiments in which completeness of aspiration of the cerebellum was verified anatomically at autopsy. The experimental results were subjected to statistical analysis.

EXPERIMENTAL RESULTS

Seizure discharges were observed to appear 6 min after application of strychnine to the cortex of the sigmoid gyrus. During the next 5-8 min the amplitude of the potentials increased to 1.5-2.5 mV. The frequency of spike generation during this period was 10-40 spikes/min. The amplitude–frequency characteristic curves of the seizure discharges remained unchanged for 5-10 min after the discharges had reached their maximal amplitude, after which the level of seizure activity of the focus declined gradually and disappeared completely. The mean life of the foci was 22.7 ± 2.6 min (Table 1).

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