Consequently, the results show that rats of different zoosocial rank are characterized by a definite specific pattern of central regulation of their hormonal functions during stress, mediated by the noradrenalin- and dopamine-synthesizing brain structures listed above.

In all cases when significant differences were observed between zoosocial groups, a higher catecholamine level was characteristic of the D rats. Similar results also have been obtained by other workers [3]. Since a high catecholamine level in the brain nuclei may be evidence of reduced release of this substance, it can be tentatively suggested that activity of catecholamine-synthesizing structures involved in the central regulation of hormonal functions during stress is lower in the D rats than in rats of the other ranks. Meanwhile, the possibility of differences in the level of catecholamine metabolism in rats of different zoosocial position cannot be ruled out.

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EFFECT OF COMBAT STRESS AND CAPITULATION STRESS ON RESISTANCE OF HEART MUSCLE TO EXCESS CALCIUM

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During long-term exposure to a stress situation the initial (adaptive) effect of stress, during which activation of the antioxidative system takes place [1], may be converted into a damaging effect due to the overactivation of lipases, phospholipases, and processes of free-radical oxidation by catecholamines [5]. This has been proved mainly in relation to combat stress, the type most frequently observed under real conditions. In emotional-painful stress, as in [9], during which the rat unsuccessfully, but persistently, realizes an electric shock avoidance reflex, exposure for 1 h leads to an increase in the force and rate of contraction, and to an even greater degree, of relaxation, of the isolated heart, but also to an increase in its resistance to hypoxia. Exposure to stress for 6 h induces the directly opposite effect. This rule was discovered in a study of the contractile function of isolated hearts of animals under isovolumic, aerobic conditions, and in the presence of a normal Ca^{2+} concentration [4].

The aims of the present investigation were, first, to discover how the adaptive effect of stress changes into a damaging effect relative to resistance of the isolated heart to the contracture-inducing action of high Ca^{2+} concentrations, and second,

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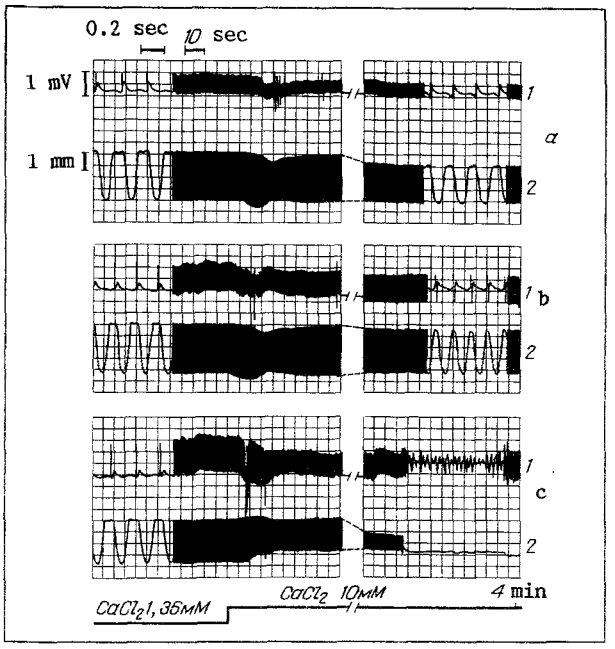

Fig. 1. Effect of emotional-painful stress of varied duration on mechanical and electrical activity of the isolated rat heart under the influence of excessive Ca^{2+} concentrations. 1) ECG, 2) amplitude of isotonic contraction (upper part of trace corresponds to level of diastolic relaxation). a) Control, b) stress for 1 h, c) stress for 6 h. Time of changing normal solution for one with high calcium concentration indicated by a step.

TABLE 1. Effect of Emotional-Painful Stress (EPS) of Varied Duration on Parameters of Contractility, Arrhythmia, and Contracture of Isolated Heart with an Increase in the Ca^{2+} Concentration in the Perfusion Solution

Parameter	Experimental group		
	control (n=8)	EPS, h	
		1	6
Heart rate, beats/min	274±8	279±13	266±8
Amplitude of shortening, mm	2,70±0,09	3,06±0,04**	2,81±0,10
Rate of contraction, mm/sec	56,8±2,4	62,0±1,4	60,2±2,2
Rate of relaxation, mm/sec	62,4±2,8	71,8±1,6*	65,6±2,8
Degree of contracture, % of initial amplitude of contractions	29,1±7,2	2,1±1,7**	46,0±14,0
Area of contracture, % of area of contractions	13,2±2,7	3,1±0,9**	19,1±5,9
Extrasystoles	10±1	2±1**	15±2*
Ventricular fibrillation, number of cases	0	0	4*

Legend. Here and in Table 2, significance of differences from control: $*p < 0.05$, $**p < 0.01$.

using the same method, to compare the cardiotropic effects of combat stress and capitulation stress caused by immobilization of the animals.

TABLE 2. Effect of Immobilization Stress of Varied Duration on Parameters of Contractility, Arrhythmia, and Contracture of Isolated Heart in Response to an Increase in Ca^{2+} Concentration in Perfusion Solution

Parameter	Experimental group		
	control (n=15)	immobilization stress, h	
		(n=11)	(n=8)
Heart rate, beats/min	274±8	269±8	272±6
Amplitude of shortening, mm	2,60±0,08	2,07±0,16**	2,66±0,14
Rate of contraction, mm/sec	51,0±2,8	42,8±3,4	54,6±3,6
Rate of relaxation, mm/sec	57,6±3,0	46,8±4,2*	64,6±3,8
Degree of contracture, % of initial amplitude of contractions	21,0±4,2	59,2±17,1*	18,3±6,0
Area of contracture, % of area of contractions	10,0±1,8	35,8±10,1*	9,4±3,9
Extrasystoles, number	5±2	7±3	3±2
Ventricular fibrillation, number of cases	0	6**	0

EXPERIMENTAL METHOD

Experiments were carried out on male Wistar rats weighing 200-250 g. Combat stress was induced in the form of emotional-painful stress as in [9], capitulation stress by immobilizing the animal by all its limbs in the supine position. Both types of stress were used in two variants: short-term, for 1-2 h, and long-term, for 6 h. Thus altogether there were six series of experiments with 8-15 animals in each series. The rats were used in the experiments 2 h after the end of exposure to stress. The method used to study cardiac function in all series of experiments was as follows. The animals were heparinized (200 U/100 g, intraperitoneally), anesthetized with pentobarbital (50 mg/kg, intraperitoneally), after which the heart was quickly removed and placed in a Langendorff perfusion system. Standard Krebs–Henseleit solution (glucose 11 mM) was used for perfusion. The solution was aerated with a gas mixture of 95% O_2 and 5% CO_2 at 37°C; pH was kept between 7.3 and 7.4. The perfusion pressure was 9.5 kPa (97 cm water). Mechanical activity of the isolated heart was assessed with the aid of a TD-112S isotonic transducer, which recorded apicobasal shortening of the heart. The degree of contracture was estimated as the absolute change in apicobasal length of the heart during diastole relative to its length at rest, measured at the end of the period of stabilization. The ECG and mechanical activity were recorded by specialized modules of the RM-6000 polygraph and VC-9 oscilloscope (Nihon Kohden, Japan). One electrode for recording the ECG was applied to the aorta at the base of the heart, the other to the left ventricle. After stabilization of the isolated heart for 20 min, the coronary flow and heart rate were measured under retrograde perfusion conditions and the principal parameters of mechanical and electrical activity were recorded: the amplitude and rate of contraction, the rate of relaxation, and the ECG. Calcium overloading was then produced by raising the $CaCl_2$ concentration in the solution from 1.36 to 10 mM, i.e., by 7.3 times, and keeping this high Ca^{2+} concentration for 4 min. In the course of the experiment the amplitude, rate of contraction, rate of relaxation, and changes arising in response to calcium overloading, i.e., the maximal value of contracture, the area of contracture, number of extrasystoles, and number of cases of fibrillation of the heart, were measured. The degree of contracture was expressed as a percentage, taking the amplitude of contractions before the Ca^{2+} concentration was increased as 100%. The area of contracture was determined planimetrically and also expressed as a percentage, taking the area of contraction for calcium overloading as 100%. The results were subjected to statistical analysis by Student's test and by Fisher's exact test.

EXPERIMENTAL RESULTS

The traces in Fig. 1 characterize one typical experiment; when they are interpreted it must be recalled that the top fragment of the trace of mechanical activity corresponds to maximal relaxation, the bottom fragment to maximal contraction of the heart, working under isotonic conditions. With a normal Ca^{2+} concentration, combat stress for 1 h induced a small increase in the amplitude and velocities of contraction and relaxation, which could not be detected after stress lasting 6 h. The main difference between combat stress for 1 h and 6 h was observed in the case of Ca^{2+} overloading. After stress for 1 h contracture and extrasystoles were virtually absent unlike in the control, and fibrillation of the heart likewise was not observed. After 6 h of stress, on the other hand, the degree and area of contracture were approximately twice as great as in the control, numerous extrasystoles occurred, and at the 3rd minute of action of Ca^{2+}, irreversible fibrillation of the heart developed. The quantitative results of these experiments are shown in Table 1.

Table 1 shows that short-term (1 h) combat stress reduced the maximal value of hypercalcium contracture 15-fold, and its area fivefold compared with the control, i.e., it virtually abolished this phenomenon or it ceased to be significant; the extrasystoles found in the control also disappeared. Exposure to stress for 6 h led to the opposite result: the degree of contracture increased by almost 60% and its area by 45%. The number of extrasystoles was increased, and fibrillation and cardiac arrest developed in half of the animals at the 3rd to 4th minutes of action of the hypercalcium solution, although this was never observed in the control or in short-term exposure to stress.

This result is unequivocal evidence that short-term combat stress increases the resistance of the heart muscle to Ca^{2+} overloading, on account of adaptive effects [6] (more especially on account of activation of the Ca-pumps of the sarcoplasmic reticulum and sarcolemma) whereas long-term stress leads to the opposite result on account of damaging changes: efficiency of membrane mechanisms of Ca^{2+} transport is reduced and, ultimately, the resistance of the heart to the contracture- and arrhythmia-inducing action of an excess of Ca^{2+} is weakened. This state of affairs, in agreement with the results of a previous study [4], is valid, however, only for combat stress, when animals, finding themselves in a threatening or even damaging situation, perform active responses in the search for escape. Under immobilization stress and capitulation conditions, when the search for an escape is completely impossible, the time course of the events in the heart is in many respects opposite to that observed during EPS. It follows from the data in Table 2 that hearts taken from animals after 2 h of exposure to immobilization stress, and in the presence of physiological Ca^{2+} concentrations, responded by a significant decrease in the amplitude of contraction and the rates of contraction and relaxation. With a 7.3-fold increase in the Ca^{2+} concentration the maximal degree of contracture, and also its area, were increased threefold in these animals compared with the control. Finally, in more than half of the animals, at the 4th minute of perfusion with the hypercalcium solution, ventricular fibrillation developed.

It was thus proved conclusively that after 2 h of capitulation stress the membrane mechanisms of calcium transport in the cardiomyocytes were disturbed sufficiently deeply, and, correspondingly, the resistance of the heart to the arrhythmia- and contracture-inducing action of an excess of calcium was reduced.

As the results in Table 2 show, hearts removed 2 h after a 6-h period of immobilization stress were indistinguishable from the control. This, of course, does not mean that all traces of damage in them had disappeared, but it does show that in animals capitulating long before the heart was investigated, the decrease in resistance to the arrhythmia- and contracture-inducing action of an excess of calcium had been abolished.

The mechanism of this urgency adaptation to enforced capitulation and the more general question of the neurohormonal, cellular, and molecular mechanisms of this difference which we found between combat stress and capitulation stress are not clear and require further study. One possibility is that during long-term capitulation stress the initial rise in blood levels of first-line stress hormones, which, together with catecholamines and corticosteroids, also includes parathormone, which raises the blood calcium concentration [8], gives way, during continuation of immobilization, to the formation of a different hormonal spectrum, for example, an increase in the blood thyrocalcitonin level, causing the plasma calcium concentration to fall [2, 3]. Of course, there are other possibilities.

Thus the structure of the stress reaction and, correspondingly, its effects in target organs are not standard, as Selye [7] postulated some time ago but, conversely, they are strongly dependent on the quality of the stress situation to which the body is exposed.

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