12. I. Namura, P. Douillet, S. E. Sun, et al., Eur. J. Pharmacol., 136, 31 (1987).
13. T. Tadano, N. Satoh, I. Sakuma, et al., Life Sci., 40, 1309 (1987).

DETERMINATION OF LEADING FACTORS IN PATHOGENESIS OF ALCOHOL CARDIOMYOPATHY

V. P. Nuzhnyi, I. G. Zabvirova,
A. Kh. Abdrashitov, and A. E. Uspenskii

UDC 616.12-02:[615.917:547.262]-092.9

KEY WORDS: ethanol; acetaldehyde; acetone; heart; rats

The aim of this investigation was to elucidate the leading mechanisms of damage to the rat heart during forced alcoholization.

EXPERIMENTAL METHOD

Experiments were carried out on noninbred male albino rats weighing 300-390 g, receiving dry food and water ad lib. Ethanol was injected into the stomach in the form of a 25% solution twice a day (at 9 a.m. and 9 p.m.) for 5.5 days. The doses of ethanol varied from 2 to 5 g/kg individually depending on the state of the animals and the time of routine injection [1]. When signs of intoxication were present, and allowing for their severity, the dose of ethanol was reduced. On the 4th day, 2 and 12 h after the 8th injection of ethanol, concentrations of ethanol, acetaldehyde, and acetone were determined [6] in blood taken from the caudal vessels. Mean 24-hourly concentrations of these compounds in the blood, the total dose of ethanol received by the rats during the experiment, and the index of tolerance of the rats to ethanol (ITE) were calculated, the latter by the formula $(A - 33)/(55 - 33)$, where A denotes the total dose of ethanol given in 5.5 days, (in g/kg), 33 is the conventional minimum total dose of ethanol (in g/kg); 55 the conventional maximal total dose of ethanol (in g/kg). The heart was removed from rats anesthetized with hexobarbital with the addition of heparin, 2-8 h after the last injection of ethanol, and perfused with Krebs–Henseleit solution containing 1% gelatinol, under a pressure of 70 mm Hg [5]. After 10 min of continuous perfusion the heart was switched to reperfusion with a volume of 35 ml of recycling solution, and this continued for 30 min. The peak systolic pressure (PSP) in the left ventricle, its components, the diastolic pressure (DP), the heart rate (HR) and the coronary flow (CF) were recorded. The pressures were measured by means of a "Statham" electromanometer, connected to a small latex balloon, inserted through an incision in the auricle of the left atrium into the left ventricle of the heart. The rate of contraction (RCH) and relaxation (RRH) of the heart and the tension time index (TTI) of the myocardium were calculated by the formula: $PSP \cdot HR \cdot T/1000$, where T denotes the tension time of the left ventricle. Creatine phosphokinase (CPK) activity was determined in the perfusion fluid [2]. At the end of perfusion the heart was dried to constant weight. Hearts of intact animals served as the controls. Student's test and Spearman's rank correlation method were used for the statistical analysis.

EXPERIMENTAL RESULTS

It will be clear from Table 1 that the method of alcoholization used in the investigation ensures a high, constant blood ethanol level and the appearance of acetaldehyde and acetone in the rat's blood. The mean 24-hourly acetaldehyde concentration correlated with the corresponding value of the blood ethanol concentration ($\rho = +0.572$, $p < 0.05$). ITE varied in different individuals from 0.32 to 0.82. Negative correlation was found between ITE and the residual blood ethanol concentration toward the time of the next routine injection ($\rho = -0.491$, $p < 0.05$). This agreement between individual doses of ethanol received by the rats on the 3rd day of the experiment and the mean-24-hourly levels ($\rho = +0.684$, $p < 0.01$)

Laboratory of Toxicology, All-Union Research Center for Medico-Biological Problems of Drug Addiction, Ministry of Health of the USSR, Moscow. (Presented by Academician of the Academy of Medical Sciences of the USSR A. V. Val'dman.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 107, No. 2, pp. 150-152, February, 1988. Original article submitted July 21, 1987.

TABLE 1. Concentrations (g/liter) of Ethanol, Acetaldehyde, and Acetone in Blood of 20 Rats 2 and 12 h, and Mean 24-Hourly Level, after Routine (8th) Injection of Ethanol

Dose of ethanol received by rats during previous 24 h, g/kg	Ethanol			Acetaldehyde			Acetone	
	after 2 h	after 12 h	mean	after 2 h	after 12 h	mean	after 2 h	after 12 h
7,9±0,2	4,43±0,26	1,22±0,23	2,88±0,22	0,0067±0,0007	0,0028±0,0005	0,0047±0,0004	0,018±0,002	0,019±0,002

TABLE 2. Parameters of Contractile Function and Outflow of CPK from Isolated Heart of Rats Subjected to Forced Alcoholization

Groups of rats	No. of observations	HR, beats/min	PSP, mm Hg	TTI; (mm Hg/min)·1000	DP, mm Hg	RCH, mm Hg/sec	RRH, mm Hg/sec	CF, ml/min	Outflow of CPK, U/g dry tissue
Control	8	237±6	130±4	2,04±0,08	7±1	1950±41	1365±46	19,2±1,7	0,322±0,058
Experimental	20	163±11**	155±5**	1,84±0,10	10±1*	2033±62	1174±34*	12,4±0,5**	1,854±0,225**

Legend. *$p < 0.05$, **$p < 0.001$ compared with control.

TABLE 3. Correlation between Blood Levels of Ethanol, Acetaldehyde, and Acetone in Rats Subjected to Forced Alcoholization and Parameters of Activity of the Isolated Heart (Spearman's Rank Correlation Coefficients – ρ)

Parameters compared	HR	PSP	TTI	RCH	RRH	CF	Rate of outflow of CPK
Blood levels after routine (8th) injection of ethanol							
Ethanol: after 2 h	–0,497	—	—	—	—	—	+0,452
After 12 h	—	—	—	—	—	—	—
Average	—	—	—	—	—	—	—
Acetaldehyde: after 2 h	—	—	—	—	–0,454	—	—
After 12 h	—	—	—	—	—	—	—
Average	—	–0,495	–0,482	–0,458	–0,469	—	—
Acetone (average)	—	—	—	—	—	—	—

Legend. In columns in which a coefficient of correlation is shown $p < 0.05$. A dash in a column signifies $\rho < 0.440$, with $p > 0.05$.

reliably showed that the concentrations of ethanol, acetaldehyde, and acetone on the 4th day of the experiment reflect sufficiently completely individual differences in the level of intoxication of the animals throughout the experiment.

Forced alcoholization led to a marked fall of HR, RCH, and CF and an increase in PSP and DP of the isolated heart (Table 2). The outflow of CPK from the heart was increased by 5.7 times. Negative correlation was found between HR and PSP ($\rho = -0.443$, $p < 0.05$). Positive correlation was found between CP and RCH ($\rho = +0.545$, $p < 0.05$).

Comparison of the severity of intoxication and parameters of cardiac activity (Table 3) revealed negative correlation between the mean 24-hourly acetaldehyde concentration, on the one hand, and PSP, TTI, RCH, and RRH. The maximal ethanol concentration in the blood correlated negatively with HR and positively with the rate of outflow of CPK. The results of the investigation revealed positive correlation between disturbances of the contractile function of the heart and the blood acetaldehyde concentration of the animals in the course of alcoholization. This contradicts the view that the direct membranotropic action of ethanol plays the leading role in the pathogenesis of alcohol cardiomyopathy [10]. The character of the disturbances of cardiac activity indicates that acetaldehyde affects ionic permeability of the cell membranes and the energy supply to the myocardium [3]. Meanwhile inhibition of the pacemaker function of the heart is evidently due to the direct action of ethanol at the height of its concentration. Elevation of PSP can be interpreted as an adaptive reaction aimed at maintaining the pumping function of the heart in response to the fall of HR. The

observed fall of CF is not directly connected with the toxic effect of ethanol and acetaldehyde on the vessels.

The existence of positive correlation between the blood ethanol concentration of the rate 2 h after routine injection of ethanol and the rate of outflow of CPK from the heart indicates that ethanol, at the height of its concentration, has a damaging action on cardiomyocytes and on their sarcolemma. It also follows from the results of this investigation that the severity of the cardiac disturbances in rats with high and low tolerance to ethanol was identical, whereas acetonemia, a manifestation of alcohol-induced ketosis, has no direct toxic action on the heart.

The results shed light on the causes of lack of success in the simulation of alcohol cardiomyopathy by the use of semivoluntary methods of alcoholization of animals [7-9]. In such cases acetaldehyde does not accumulate and the highest blood ethanol concentrations do not exceed 2 g/liter [1, 4].

The leading factors in heart damage during forced alcoholization of rats are thus the accumulation of acetaldehyde in the body and the presence of periodically occurring high blood ethanol concentrations.

LITERATURE CITED

1. A. Kh. Abdrashitov, V. P. Listvina, V. P. Nuzhnyi, and A. A. Uspenskii, Farmakol. Toksikol., 44, No. 6, 94 (1983).
2. F. B. Levin and I. M. Yakobson, Lab. Delo, No. 4, 223 (1979).
3. F. Z. Meerson, Pathogenesis and Prevention of Stress-Induced and Ischemic Heart Damage [in Russian], Moscow (1984).
4. V. P. Nuzhnyi, A. Kh. Abdrashitov, V. P. Listvina, and A. E. Uspenskii, Farmakol. Toksikol., 49, No. 6, 96 (1986).
5. V. P. Nuzhnyi, E. B. Tezikov, E. V. Savitskaya, and A. I. Igryumov, Byull. Éksp. Biol. Med., 101, No. 5, 575 (1986).
6. A. E. Uspenskii, V. P. Nuzhnyi, A. Kh. Abdrashitov, and V. P. Listvina, Biological Bases of Alcoholism [in Russian], Moscow (1984), pp. 86-92.
7. J. L. Hall and D. T. Rowlands, Am. J. Pathol., 60, No. 1, 153 (1970).
8. A. Hepp, T. Rudolph, and K. Kochsiek, Basic Res. Cardiol., 79, No. 2, 230 (1984).
9. T. Mattfeldt, G. Mali, and B. Volk, J. Mol. Cell. Cardiol., 12, No. 11, 1311 (1980).
10. E. Rubin, Fed. Proc., 41, No. 8, 2460 (1982).

ABOLITION OF DISTURBANCES OF ELECTRICAL STABILITY OF THE HEART IN POSTINFARCTION CARDIOSCLEROSIS BY A FACTOR INDUCING GABA ACCUMULATION IN THE BRAIN

N. A. Abdikaliev, E. V. Shabunina, and F. Z. Meerson

UDC 616.127-005.8-036.8:616.12-004.6]-07:616.12-009.3-073.97-02:615.213

KEY WORDS: sodium valproate, postinfarction cardiosclerosis, fibrillation of the heart

Recent investigations have shown that adaptation to short-term stress not only limits many kinds of stress-induced damage [5], but also prevents arrhythmia and fibrillation of the heart associated with acute ischemia, myocardial infarction, and disturbance of the electrical stability of the heart in postinfarction cardiosclerosis [4]. Studies of the protective effect of adaptation have shown that it is based on activation of the stress-limiting system of the body and, in particular, of the GABA-ergic system, which is manifested as marked accumulation of GABA in the brain. In complete agreement with this it has been shown that injection of a chemical factor, inducing GABA accumulation in the brain, namely sodium

Research Institute of General Pathology and Pathological Physiology, Academy of Medical Sciences of the USSR, Moscow.

(Presented by Academician of the Academy of Medical Sciences of the USSR I. K. Shkhvatsabaya.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 107, No. 2, pp. 152-154, February, 1989. Original article submitted January 20, 1988.