

# Effects of Acute Administration of Ethanol on Experimental Arrhythmia

Yang Gao<sup>1</sup>, Peng Li<sup>1</sup>, Li-Xia Ma<sup>2</sup>, Ke-Xin Du<sup>3</sup>, Xing-Hui Wang<sup>3</sup>, Meng-Jie Tang<sup>1</sup>,  
Hui-Kang He<sup>1</sup>, Xiao-Jiang Yu<sup>1</sup>, Wei-Jin Zang<sup>1</sup>, and Hao Hu<sup>1</sup>

<sup>1</sup>Department of Pharmacology, College of Medicine, Xi'an Jiaotong University

<sup>2</sup>Department of Forensic Sciences, College of Medicine, Xi'an Jiaotong University  
and

<sup>3</sup>Center for Functional Experiment Teaching, College of Medicine, Xi'an Jiaotong University  
Xi'an 710061, Shaanxi, People's Republic of China

## Abstract

Many studies have shown that the relationship between alcohol consumption and most cardiovascular diseases is U-shaped, with nondrinkers and heavier drinkers having higher risks than moderate drinkers. However, the association between cardiac arrhythmias and acute alcohol consumption is not well understood. We set up several experimental arrhythmia animal models to examine the effects of acute administration of ethanol on arrhythmia. The results showed 0.4, 0.8 and 1.6 g/kg ethanol consumption obviously delayed the onset time of atrial fibrillation (AF) ( $P < 0.05$  or  $P < 0.01$ ) and increased the survival rates on acetylcholine-CaCl<sub>2</sub>-induced AF in mice. Ethanol (0.4, 0.8 and 1.6 g/kg) consumption significantly delayed the onset time of ventricular tachycardia (VT), ventricular fibrillation (VF) and cardiac arrest (CA) ( $P < 0.01$ ), and 0.4 and 0.8 g/kg ethanol consumption increased the survival rates on CaCl<sub>2</sub>-induced arrhythmia in rats. Ethanol (0.4 g/kg) essentially increased the cumulative dosage of aconitine required to CA ( $P < 0.05$ ), and 0.8 g/kg, 1.6 g/kg ethanol reduced the cumulative aconitine dosage to induce VT, VF and CA ( $P < 0.05$  or  $P < 0.01$ ) on aconitine-induced arrhythmia in rats. Ethanol (0.4, 0.8 and 1.6 g/kg) consumption remarkably increased the cumulative dosage of deslanoside to induce ventricular premature contraction ( $P < 0.01$ ) on deslanoside-induced arrhythmia in guinea pigs. Collectively, our results indicate that low concentrations of ethanol had anti-arrhythmic effect on experimental arrhythmia, and high concentrations of ethanol may aggravated the occurrence of experimental arrhythmia.

**Key Words:** atrial arrhythmia, ethanol, ventricular arrhythmia

## Introduction

Many studies have shown an inverse relationship between increasing alcohol consumption and a decrease in risk of cardiovascular disease (4, 16). Although acute alcohol intoxication has long been implicated in the development of atrial fibrillation (AF) in healthy individuals, the relationship of the full range of alcohol consumption with risk of incidence of AF is less certain (14).

Some studies have found a higher risk of AF

among heavier drinkers than in moderate drinkers (8, 13). However, a large number of epidemiological studies reported that ethanol was not associated with occurrences of AF (3, 5). While our current knowledge indicates that long-term heavy alcohol consumption may promote structural myocardial changes that can be pro-arrhythmic, it is still debatable whether and how acute ethanol ingestion favor the genesis of atrial arrhythmias, particularly in patients without preexisting heart disease. Moreover, limited experimental studies have suggested that the influence of ethanol on elec-

trical stimulation-induced arrhythmias have also shown contradictory results, with the drug either increasing or decreasing susceptibility to and the duration of AF in animal models (1, 15). Thus, the association between atrial arrhythmias and alcohol consumption is not well understood. Furthermore, the effect of ethanol on ventricular arrhythmia is also uncertain.

This controversy is, to some extent, fueled by the fact that the acute electrophysiologic effects of alcohol *in vivo* have not been unequivocally characterized. To more fully address the prospective association of alcohol consumption and risk of arrhythmias, we designed several experimental arrhythmia animal models to investigate the effects of acute administration of different concentrations of ethanol on arrhythmia.

## Materials and Methods

### *Animals and Reagents*

This study was performed in accordance with the guidelines for animal handling and experimentation established at Xi'an Jiaotong University, China. Male ICR mice (18-22 g), Sprague-Dawley rats (250-300 g) and guinea pigs (300-350 g) were obtained from the Laboratorial Animal Center of Xi'an Jiaotong University. All experimental procedures and protocols were reviewed and approved by the Animal Care and Use Committee of Medical College, Xi'an Jiaotong University. Acetylcholine and aconitine were purchased from Sigma-Aldrich (St. Louis, MO, USA), amiodarone and verapamil were purchased from Harvest Pharmaceutical Co. Ltd. (Shanghai, PRC), ethanol, CaCl<sub>2</sub> and urethane were purchased from Xi'an Chemical Agent Factory (Xi'an, Shaanxi, PRC). Animals were anaesthetized intraperitoneally with urethane (0.12 g/kg), and trachea intubation was made for each animal to assist ventilation. The lead II ECG was recorded by BL-420E+ data acquisition and analysis system (Chengdu, Sichuan, PRC). Drugs were administered *via* the caudalis vein in mice, the sublingual vein in rats, and the jugular vein in guinea pigs.

### *Acetylcholine-CaCl<sub>2</sub>-Induced AF in Mice*

Fifty mice were randomly divided into five groups that were the normal saline (10 ml/kg), amiodarone (5 mg/kg), 5% ethanol (0.4 g/kg), 10% ethanol (0.8 g/kg) and 20% ethanol (1.6 g/kg) groups. Five minutes after treatment of amiodarone or ethanol, acetylcholine-CaCl<sub>2</sub> (consisting of 6 mg CaCl<sub>2</sub> and 25 µg acetylcholine per ml) was administered intravenously in 5 s. The onset time and duration of AF (f waves) or atrial flutter (F waves) were recorded, and the survival rates of different groups animals were calculated.

### *CaCl<sub>2</sub>-Induced Ventricular Arrhythmia in Rats*

Forty rats were randomly divided into five groups that were the normal saline (10 ml/kg), verapamil (1 mg/kg), 5% ethanol (0.4 g/kg), 10% ethanol (0.8 g/kg), and 20% ethanol (1.6 g/kg) groups. Five minutes after treatment of verapamil or ethanol, 3.5% CaCl<sub>2</sub> (140 mg/kg) was administered intravenously in 10 s. The onset time of ventricular premature contraction (VPC), ventricular tachycardia (VT), ventricular fibrillation (VF) and cardiac arrest (CA) was recorded, and the survival rates of different groups of animals were calculated.

### *Aconitine-Induced Ventricular Arrhythmia in Rats*

Forty rats were randomly divided into five groups that were the normal saline (10 ml/kg), amiodarone (5 mg/kg), 5% ethanol (0.4 g/kg), 10% ethanol (0.8 g/kg) and 20% ethanol (1.6 g/kg) groups. Five minutes after treatment of amiodarone or ethanol, 0.001% aconitine was administered at a rate of 2 µg/min using an infusion pump. The onset time of VPC, VT, VF and CA was recorded, and the cumulative dosage of aconitine in mg/kg required to induce VPC, VT, VF and CA was calculated.

### *Deslanoside-Induced Ventricular Arrhythmia in Guinea Pigs*

Forty guinea pigs were randomly divided into five groups that were the normal saline (10 ml/kg), amiodarone (5 mg/kg), 5% ethanol (0.4 g/kg), 10% ethanol (0.8 g/kg) and 20% ethanol (1.6 g/kg) groups. Five minutes after treatment of amiodarone or ethanol, 0.02% deslanoside was administered at a rate of 50 µg/min using an infusion pump. The onset time of VPC, VT, VF and CA was recorded, and the cumulative dosage of deslanoside in mg/kg required to induce VPC, VT, VF and CA was calculated.

### *Measurement of Concentration of Blood Ethanol*

Five minutes after treatment of different concentrations of ethanol, the arterial blood samples were obtained. Blood ethanol levels were determined by gas chromatography (12).

### *Statistical Analysis*

Data are presented as means ± SD (standard deviation), and n equals the number of animals studied. Statistical analysis was performed by Student's *t*-test or one-way analysis of variance followed by a least significant difference *post-hoc* test for multiple comparisons. The software used for data analysis is the

**Table 1. Effects of ethanol on acetylcholine-CaCl<sub>2</sub>-induced atrial fibrillation in mice**

Groups	Doses (g/kg)	Onset Time of AF (s)	Duration of AF (s)
Normal saline		1.50 ± 1.08	39.86 ± 8.88
Amiodarone	0.005	1.70 ± 0.48	18.30 ± 8.89**
5% Ethanol	0.4	4.30 ± 1.64**	29.70 ± 7.17*
10% Ethanol	0.8	2.90 ± 1.10*	35.70 ± 10.35
20% Ethanol	1.6	3.00 ± 1.25*	37.50 ± 11.36

In each group, n = 10, and the data shown are means ± SD. \* $P < 0.05$ , \*\* $P < 0.01$  vs. normal saline.

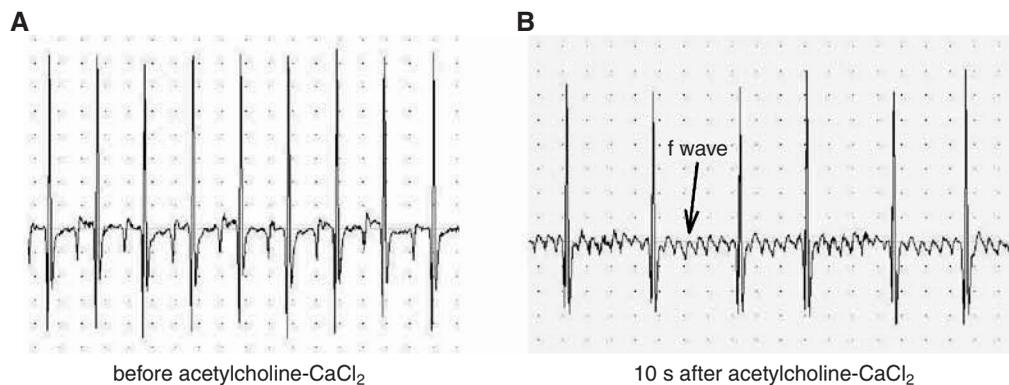


Fig. 1. Electrocardiogram of atrial fibrillation (AF) induced by acetylcholine-CaCl<sub>2</sub> in mice. (A) Normal electrocardiogram of lead II in a mouse. (B) AF (characterized as f waves) was generated in the mouse after administration of the acetylcholine-CaCl<sub>2</sub> mixture.

Statistical Package for the Social Sciences (SPSS) of windows version 12.0.

## Results

### *Effects of Ethanol on Acetylcholine-CaCl<sub>2</sub>-Induced AF in Mice*

Five minutes after treatment of 0.4, 0.8 and 1.6 g/kg ethanol, the concentrations of blood ethanol in the treated mice were  $11.57 \pm 0.07$ ,  $22.03 \pm 0.75$  and  $44.07 \pm 3.85$  mM, respectively. AF (characterized as f waves) or atrial flutter (characterized as F waves) was generated in all the mice after administrating of acetylcholine-CaCl<sub>2</sub> mixture (Fig. 1). Our results showed that 0.4, 0.8 and 1.6 g/kg ethanol obviously delayed the onset time of AF ( $P < 0.05$  or  $P < 0.01$ ), and 0.4 g/kg ethanol and amiodarone also remarkably shortened the duration of AF ( $P < 0.05$  or  $P < 0.01$ ) (Table 1 and Fig. 1).

The administration of acetylcholine-CaCl<sub>2</sub> mixture results in death in some animals. The incidence of mortality was 30% (3 of 10 animals) in the normal saline group, 20% (2 of 10 animals) in the 1.6 g/kg ethanol group, and all animals survived in the 0.4 and 0.8 g/kg ethanol and amiodarone groups (Fig. 2).

### *Effects of Ethanol on CaCl<sub>2</sub>-Induced Ventricular Arrhythmia in Rats*

Five minutes after treatment of 0.4, 0.8 and 1.6 g/kg ethanol, the concentrations of blood ethanol in the experimental rats were  $10.47 \pm 1.04$ ,  $19.40 \pm 1.64$  and  $48.20 \pm 1.83$  mM, respectively. Intravenous injection of CaCl<sub>2</sub> to rats produced disturbances of cardiac rhythm including VPC, VT, VF or CA (Fig. 3). Our data also indicated that intravenous administration of 0.4, 0.8 and 1.6 g/kg ethanol and verapamil significantly delayed the onset time of VT, VF and CA ( $P < 0.01$ ). Additionally, 0.4 g/kg ethanol and verapamil also remarkably delayed the onset time of VPC ( $P < 0.05$ ) (Table 2 and Fig. 3).

Administration of CaCl<sub>2</sub> caused life-threatening arrhythmia in most animals in all groups. The incidence of mortality reached 100% (8 of 8 animals) in the normal saline and 1.6 g/kg ethanol groups, 50% (4 of 8 animals) in the verapamil and 0.4 g/kg ethanol groups, and 87.5% (7 of 8 animals) in the 0.8 g/kg ethanol group (Fig. 4).

### *Effects of Ethanol on Aconitine-Induced Ventricular Arrhythmia in Rats*

Ventricular premature beats were followed by

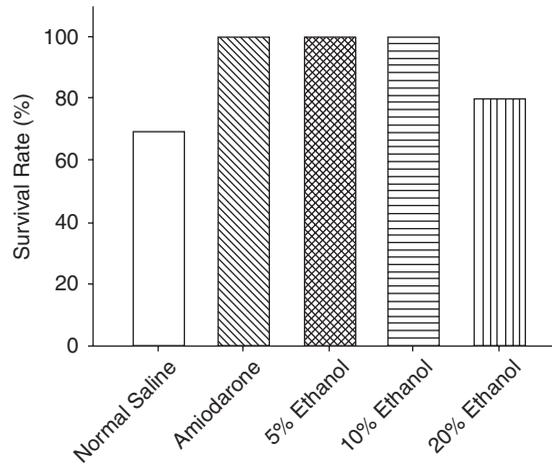


Fig. 2. Survival rates of ethanol on acetylcholine-CaCl<sub>2</sub>-induced AF in mice. The survival rate in the normal saline group was 70%, in the 1.6 g/kg ethanol group was 80%, and in the 0.4 and 0.8 g/kg ethanol and amiodarone groups was 100%.

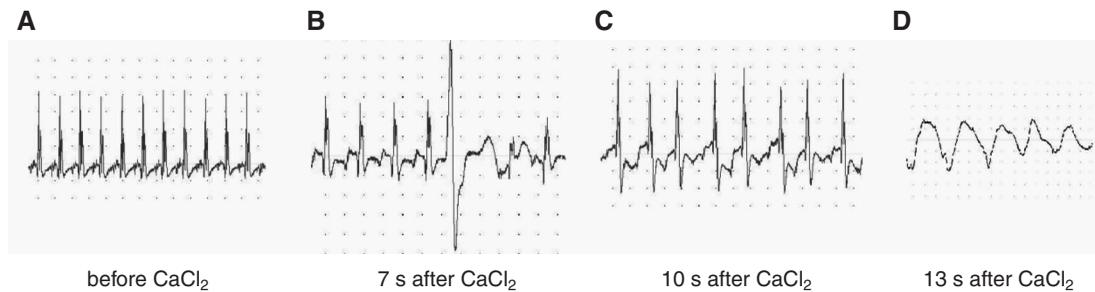


Fig. 3. Electrocardiogram of ventricular arrhythmia induced by CaCl<sub>2</sub> in rats. (A) Normal electrocardiogram of lead II in a rat. (B) VPC was generated in the rat induced by CaCl<sub>2</sub>. (C) VT was generated in the rat induced by CaCl<sub>2</sub>. (D) VF was generated in the rat induced by CaCl<sub>2</sub>.

**Table 2. Effects of ethanol on CaCl<sub>2</sub>-induced ventricular arrhythmia in rats**

Groups	Doses (g/kg)	Onset Time of VPC (s)	Onset Time of VT (s)	Onset Time of VF (s)	Onset Time of CA (s)
Normal saline		7.42 ± 1.41	10.01 ± 1.41	13.65 ± 1.97	42.65 ± 5.64
Verapamil	0.001	9.12 ± 1.56*	18.73 ± 1.89**	33.91 ± 3.69**	91.41 ± 10.44**
5% Ethanol	0.4	9.34 ± 1.71*	20.34 ± 2.26**	32.91 ± 4.43**	73.64 ± 11.72**
10% Ethanol	0.8	8.65 ± 1.58	15.56 ± 1.91**	19.16 ± 2.13**	79.67 ± 9.33**
20% Ethanol	1.6	8.54 ± 1.55	13.85 ± 1.47**	24.43 ± 2.71**	86.18 ± 9.55**

In each group, n = 8, and the data shown are means ± SD. \**P* < 0.05, \*\**P* < 0.01 vs. Normal saline.

VT and VF appearing in all treated rats after administering of aconitine. Treatment of rats with 0.4 g/kg ethanol prior to aconitine caused a delay of the arrhythmias, significantly increasing the cumulative dosage of aconitine required to CA (*P* < 0.05). However, 0.8 g/kg ethanol essentially reduced the cumulative aconitine dosage to induce VF (*P* < 0.01), and 1.6 g/kg ethanol significantly reduced the cumulative aconitine dosage to induce VT, VF and CA (*P* < 0.05 or *P* < 0.01). Intravenous injection of ami-

odaron markedly and dose-dependently increased the cumulative dosage of aconitine required to induce VPC, VT, VF and CA (*P* < 0.01) (Table 3).

#### *Effects of Ethanol on Deslanoside-Induced Ventricular Arrhythmia in Guinea Pigs*

Five minutes after treatment of 0.4, 0.8 and 1.6 g/kg ethanol, the concentrations of blood ethanol in the treated guinea pigs were 11.21 ± 1.52, 20.48 ±

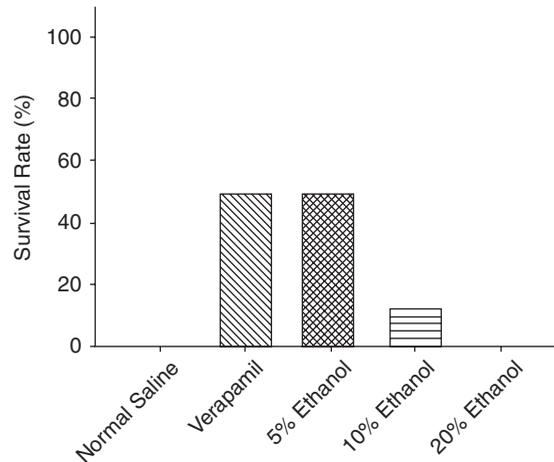


Fig. 4. Survival rates of ethanol on  $\text{CaCl}_2$ -induced ventricular arrhythmia in rats. The survival rate in the normal saline and the 1.6 g/kg ethanol groups was 0%, in the verapamil and 0.4 g/kg ethanol groups was 50%, and in the 0.8 g/kg ethanol group was 12.5%.

**Table 3. Effects of ethanol on aconitine-induced ventricular arrhythmia in rats**

Groups	Doses (g/kg)	Dosage of aconitine ( $\mu\text{g}/\text{kg}$ )			
		VPC	VT	VF	CA
Normal saline		$11.65 \pm 2.12$	$15.22 \pm 1.36$	$29.55 \pm 4.64$	$92.89 \pm 10.07$
Amiodarone	0.005	$17.15 \pm 4.19^{**}$	$22.97 \pm 6.66^{**}$	$37.59 \pm 8.00^*$	$164.98 \pm 29.66^{**}$
5% Ethanol	0.4	$12.81 \pm 3.83$	$17.87 \pm 4.75$	$34.71 \pm 7.11$	$118.63 \pm 28.63^*$
10% Ethanol	0.8	$11.20 \pm 3.03$	$14.71 \pm 3.83$	$21.67 \pm 4.44^{**}$	$83.45 \pm 17.69$
20% Ethanol	1.6	$9.91 \pm 2.75$	$12.41 \pm 2.71^*$	$18.62 \pm 3.33^{**}$	$66.87 \pm 6.90^{**}$

In each group,  $n = 8$ , and the data shown are means  $\pm$  SD. \* $P < 0.05$ , \*\* $P < 0.01$  vs. Normal saline.

2.46 and  $43.81 \pm 3.88$  mM, respectively. Intravenous injection of deslanoside resulted in disturbances of the cardiac rhythm progressing from premature ventricular to VT, VF or cardiac arrest in all guinea-pigs. Ethanol in 0.4, 0.8 and 1.6 g/kg dosages caused a remarkable increase in the cumulative dosage of deslanoside to induce VPC ( $P < 0.01$ ), but there was no obvious difference in the case of inducing VT, VF and CA ( $P > 0.05$ ) (Table 4).

### Discussion

In the present study, several drug-induced arrhythmia animal models was set up to investigate the effects of acute administration of different concentrations of ethanol on arrhythmia. The results showed that low concentrations of ethanol (0.4 g/kg) had an evidently anti-arrhythmic effect on acetylcholine- $\text{CaCl}_2$ -induced AF in mice,  $\text{CaCl}_2$  and aconitine induced ventricular arrhythmia in rats, and deslanoside-induced ventricular arrhythmia in guinea pigs. However, high concentrations of ethanol (1.6 g/kg) aggravated the occurrence of

aconitine-induced ventricular arrhythmia in rats.

It is well established that a tonic increase in the availability of the atrial muscarinic  $\text{K}^+$  channels, by infusion of a low-dose of cholinergic or adenosine receptor agonists, promotes the genesis of AF (17). On the other hand, infusion of  $\text{CaCl}_2$  leads to hypercalcemia and increase in  $\text{Ca}^{2+}$  inflow, which results in the increase of autorhythmicity of the myocardium to produce atrial or ventricular arrhythmia (2). Our data showed that 0.4, 0.8 and 1.6 g/kg ethanol not only delayed the onset time of AF on acetylcholine- $\text{CaCl}_2$ -induced AF in mice, ethanol at these concentrations also delayed the onset time of VT, VF and CA on  $\text{CaCl}_2$ -induced ventricular arrhythmia in rats. Additionally, the survival rates in all the ethanol groups were higher than those of the control group. These results were consistent with some previous studies, which confirmed an antiarrhythmic effect of ethanol (7, 15). The above results could be partially explained by another report indicating that ethanol could inhibit  $\text{Ca}^{2+}$  channel currents (9). However, little information is available on the effects of ethanol on acetylcholine

**Table 4. Effects of ethanol on deslanoside-induced ventricular arrhythmia in guinea pigs**

Groups	Doses (g/kg)	Dosage of deslanoside (mg/kg)			
		VPC	VT	VF	CA
Normal saline		0.78 ± 0.18	1.26 ± 0.32	1.81 ± 0.38	2.27 ± 0.48
Amiodarone	0.005	2.18 ± 0.10**	2.53 ± 0.16**	3.15 ± 0.19**	3.80 ± 0.26**
5% Ethanol	0.4	1.16 ± 0.15**	1.54 ± 0.19	1.94 ± 0.27	2.62 ± 0.30
10% Ethanol	0.8	1.09 ± 0.09**	1.51 ± 0.20	1.71 ± 0.22	2.12 ± 0.26
20% Ethanol	1.6	1.03 ± 0.04**	1.51 ± 0.08	1.71 ± 0.11	2.08 ± 0.14

In each group, n = 8, and the data shown are means ± SD. \**P* < 0.05, \*\**P* < 0.01 vs. Normal saline.

sensitive K<sup>+</sup> channel currents.

Aconitine, a specific Na<sup>+</sup> channel agonist able to prolong the open state of the channel, may induce intracellular Na<sup>+</sup> accumulation and intracellular Ca<sup>2+</sup> overload, which may eventually result in polymorphic ventricular arrhythmias (20). Our results indicated that there was an obvious variability of the different concentrations of ethanol on arrhythmia. An ethanol concentration of 0.4 g/kg increased the threshold dose of aconitine required to CA. However, 0.8 g/kg ethanol essentially reduced the threshold dose of aconitine to induce VF, and 1.6 g/kg ethanol reduced the threshold dose of aconitine to induce VT, VF and CA. Therefore, 0.4 g/kg ethanol showed antiarrhythmic effect on the aconitine-induced arrhythmia model, which may be due to decrease of Na<sup>+</sup> currents induced by ethanol in ventricular cardiomyocytes (10). However, more complex mechanisms might be involved in the proarrhythmic effects of high-dose ethanol on the aconitine-induced arrhythmia model, and elucidation of the effect needs more studies.

Deslanoside could elevate the intracellular sodium level to promote the reverse mode (Ca<sup>2+</sup>-entry) of the Na<sup>+</sup>/Ca<sup>2+</sup> exchanger, which could mediate ventricular arrhythmia. Guinea pig was chosen as an experimental model in this study because the Ca<sup>2+</sup>-entry/exit mechanisms in guinea pig cardiomyocytes are closely related to the human model (18). Our data showed that 0.4, 0.8 and 1.6 g/kg ethanol increased the threshold dose of deslanoside to induce VPC, but there was no obvious influence in the case of inducing VT, VF and CA. Although ethanol could inhibit the Ca<sup>2+</sup> channel currents, it seems that ethanol has little effects on the Na<sup>+</sup>/Ca<sup>2+</sup> exchanger. Hence, ethanol could not prevent increase of rhythmicity of the ventricular myocardium.

Alcohol has long been considered to be associated with different types of cardiac arrhythmias, among which the most famous case is the “holiday heart syndrome” (6). Although previous studies have suggested that alcohol consumption may induce the occurrence of AF in the so-called “holiday heart syndrome”, the association between alcohol ingestion

and AF has been challenged by several epidemiological studies (5, 8). On the other hand, some researchers pointed out that heavy drinking of alcohol was associated with increased risks of ventricular arrhythmias and sudden deaths (11, 19). But the influence of moderate drinking on ventricular arrhythmia is uncertain. Our results indicated that low concentrations of ethanol (blood concentration about 10–12 mM) had a remarkably anti-arrhythmic effect on atrial and ventricular arrhythmia, especially for atrial arrhythmia. However, high concentrations of ethanol (blood concentration about 40–50 mM) aggravated the occurrence of ventricular arrhythmia. The data obtained from four experimental arrhythmia animal models provided us with more information on the effects of ethanol on arrhythmia. Nevertheless, the exact mechanisms of these beneficial or malignant effects are unclear, thus, warranting further studies.

Although excessive alcohol consumption is associated with several negative health outcomes, more and more epidemiological and clinical investigations have shown that a low to moderate levels of alcohol intake have a definitive protective role against many cardiovascular diseases, especially on coronary heart disease and stroke. Our findings revealed that low doses of ethanol had an anti-arrhythmic effect on arrhythmia. It seems appropriate to consume alcoholic beverages in moderate amounts, which may represent a simple way to induce a cardioprotective state.

### Acknowledgments

This work was supported by the Chinese National Undergraduate Innovational Experimentation Program (2009), the Sci-tech Research and Development Project of Shaanxi Province (2008k13-04) and Guang Hua Medical Innovational Research Program of Xi'an Jiaotong University (2010).

### References

1. Anadon, M.J., Almendral, J., González, P., Zaballos, M., Delcan, J.L. and De, Guevara, J.L. Alcohol concentration determines the

- type of atrial arrhythmia induced in a porcine model of acute alcoholic intoxication. *Pacing Clin. Electrophysiol.* 19: 1962-1967, 1996.
2. Bryzgalov, A.O., Dolgikh, M.P., Sorokina, I.V., Tolstikova, T.G., Sedova, V.F. and Shkurko, O.P. Antiarrhythmic activity of 4,6-di(het)aryl-5-nitro-3,4-dihydropyrimidin-(1H)-2-ones and its effects on arterial pressure in rats. *Bioorg. Med. Chem. Lett.* 16: 1418-1420, 2006.
  3. Conen, D., Tedrow, U.B., Cook, N.R., Moorthy, M.V., Buring, J.E. and Albert, C.M. Alcohol consumption and risk of incident atrial fibrillation in women. *JAMA* 300: 2489-2496, 2008.
  4. Di Castelnuovo, A., Costanzo, S., di Giuseppe, R., de Gaetano, G. and Iacoviello, L. Alcohol consumption and cardiovascular risk: mechanisms of action and epidemiologic perspectives. *Future Cardiol.* 5: 467-477, 2009.
  5. Djoussé, L., Levy, D., Benjamin, E.J., Blease, S.J., Russ, A., Larson, M.G., D'Agostino, R.B., Wolf, P.A. and Ellison, R.C. Long-term alcohol consumption and the risk of atrial fibrillation in the Framingham Study. *Am. J. Cardiol.* 93: 710-713, 2004.
  6. Ettinger, P.O., Wu, C.F., De La Cruz, C. Jr, Weisse, A.B., Ahmed, S.S. and Regan, T.J. Arrhythmias and the "Holiday Heart": alcohol-associated cardiac rhythm disorders. *Am. Heart J.* 95: 555-562, 1978.
  7. Fenelon, G., Balbão, C.E.B., Fernandes, R., Arfelli, E., Landim, P., Ayres, O. and de Paola, A.A.V. Characterization of the acute cardiac electrophysiologic effects of ethanol in dogs. *Alcohol. Clin. Exp. Res.* 31: 1574-1580, 2007.
  8. Frost, L. and Vestergaard, P. Alcohol and risk of atrial fibrillation or flutter: a cohort study. *Arch. Intern. Med.* 164: 1993-1998, 2004.
  9. Habuchi, Y., Furukawa, T., Tanaka, H., Lu, L.L., Morikawa, J. and Yoshimura, M. Ethanol inhibition of  $Ca^{2+}$  and  $Na^+$  currents in the guinea-pig heart. *Eur. J. Pharmacol. Environ. Toxicol. Pharmacol.* 292: 143-149, 1995.
  10. Klein, G., Gardiwal, A., Schaefer, A., Panning, B. and Breitmeier, D. Effect of ethanol on cardiac single sodium channel gating. *Forensic Sci. Int.* 171: 131-135, 2007.
  11. Kloner, R.A. and Rezkalla, S.H. To drink or not to drink? That is the question. *Circulation* 116: 1306-1317, 2007.
  12. Kristoffersen, L., Stormyhr, L.E. and Smith-Kielland, A. Headspace gas chromatographic determination of ethanol: the use of factorial design to study effects of blood storage and headspace conditions on ethanol stability and acetaldehyde formation in whole blood and plasma. *Forensic Sci. Int.* 161: 151-157, 2006.
  13. Mukamal, K.J., Psaty, B.M., Rautaharju, P.M., Furberg, C.D., Kuller, L.H., Mittleman, M.A., Gottdiener, J.S. and Siscovick, D.S. Alcohol consumption and risk and prognosis of atrial fibrillation among older adults: the cardiovascular health study. *Am. Heart J.* 153: 260-266, 2007.
  14. Mukamal, K.J., Tolstrup, J.S., Friberg, J., Jensen, G. and Grønbaek, M. Alcohol consumption and risk of atrial fibrillation in men and women: the copenhagen city heart study. *Circulation* 112: 1736-1742, 2005.
  15. Nguyen, T.N., Friedman, H.S. and Mokraoui, A.M.M. Effects of alcohol on experimental atrial fibrillation. *Alcohol. Clin. Exp. Res.* 11: 474-476, 1987.
  16. Puddey, I.B., Rakic, V., Dimmitt, S.B. and Beilin, L.J. Influence of pattern of drinking on cardiovascular disease and cardiovascular risk factors—a review. *Addiction* 94: 649-663, 1999.
  17. Sarmast, F., Kolli, A., Zaitsev, A., Parisian, K., Dhamoon, A.S., Guha, P.K., Warren, M., Anumonwo, J.M.B., Taffet, S.M., Berenfeld, O. and Jalife, J. Cholinergic atrial fibrillation:  $I_{K_{ACH}}$  gradients determine unequal left/right atrial frequencies and rotor dynamics. *Cardiovasc. Res.* 59: 863-873, 2003.
  18. Shpak, B., Gofman, Y., Shpak, C., Hiller, R., Boyman, L. and Khananshvili, D. Effects of purified endogenous inhibitor of the  $Na^+/Ca^{2+}$  exchanger on ouabain-induced arrhythmias in the atria and ventricle strips of guinea pig. *Eur. J. Pharmacol.* 553: 196-204, 2006.
  19. Wannamethee, G. and Shaper, A.G. Alcohol and sudden cardiac death. *Brit. Heart J.* 68: 443-448, 1992.
  20. Zhou, S.S., Yang, J., Li, Y.Q., Zhao, L.Y., Xu, M. and Ding, Y.F. Effect of  $Cl^-$  channel blockers on aconitine-induced arrhythmias in rat heart. *Exp. Physiol.* 90: 865-872, 2005.