($P<0.01$, $n=8$)；Hill 系数 n 降低了 0.29 ($P<0.01$, $n=8$)；2) 在保留了 SR 的标本上，MCI-154 不能引起 SR 内 Ca^{2+} 释放，并对咖啡因引起的挛缩幅值无显著影响($P>0.05$)。结论：MCI-154 直接增强心肌收缩蛋白的 Ca^{2+} 敏感性，但对 SR 的 Ca^{2+} 释放无明显作用。

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Altered α_1-adrenoceptor subtypes mediated cardiac function after treatment of propranolol to rats

LI Wan, ZHANG Ying, WANG Xiao-Liang (*Department of Pharmacology, Institute of Materia Medica, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing 100050, China*)

KEY WORDS papillary muscles; heart atrium; myocardial contraction; heart rate; alpha-1 adrenergic receptors; propranolol; phenylephrine; clonidine; urapidil; carbachol

AIM: To study inotropic and chronotropic effects mediated by α_{1A}- and α_{1B}-adrenoceptors after 5-d propranolol (Pro) treatment. **METHODS**: The positive inotropic and chronotropic effects mediated by α_{1A} and α_{1B} subtypes were determined on isolated left ventricular papillary muscles and right atrium in Pro- and NaCl-treated rats. **RESULTS**: The basic contractility of papillary muscles induced by phenylephrine (Phe) was 90 ± 18 mg in Pro-treated rats and 53 ± 17 mg in control group ($P<0.05$). The increment on force of contraction was 20 ± 12 mg in Pro-pretreated rats and 5 ± 5 mg in NaCl-treated rats ($P<0.05$). After preincubated with chloroethylclonidine, the increment on force of contraction was reduced in Pro-treated rats, but was not much changed in control group. Phe in presence of 5-methylurapidil induced positive inotropic effect with 13 ± 5 mg in Pro-treated group, but not in NaCl-treated rats. Under the normal and the inhibited cardiac state, the maximal increment in beat rate mediated by α_{1B} showed no difference between the Pro-treated and NaCl-treated rats. **CONCLUSION**: After chronic treatment of Pro, α_1-adrenoceptor-mediated positive inotropic effect in rat heart was improved, which was mainly induced by stimulation of α_{1B} when β-adrenoceptors were blocked.

Myocardial α_1- and β-adrenoceptors were coexistent in hearts of various species, including rat[1]. Both adrenoceptors mediate positive inotropic and chronotropic effects. Since the β-adrenoceptors-mediated responses are "dominant," β-adrenoceptors blockade enhances the significance of α_1-adrenoceptor-mediated effects. α_1-Adrenoceptor density increases in rat heart after chronic propranolol treatment[2,3]. We demonstrated that α_{1A}- receptor density increased more pronounced than α_{1B}-adrenoceptor after chronic treatment of propranolol (Pro)[4]. The present experiment was to observe the functional alterations of positive inotropic and chronotropic effects mediated by α_{1A}- and α_{1B}-adrenoceptors after chronic treatment of Pro to rats.

MATERIALS AND METHODS

Wistar rats, ♂, 230 - 260 g, were treated with Pro (50 $mg \cdot kg^{-1}$, ip, bid) or 0.9 % NaCl solution for 5 d.

Force of contraction Papillary muscles isolated from the left ventricles of Pro- and NaCl-treated rats were attached to a stimulating electrode and suspended in 15 mL Krebs' solution: NaCl 118, KCl 4.7, $NaHCO_3$ 4.5, $MgSO_4 \cdot 7H_2O$ 0.45, KH_2PO_4 1.03, glucose 11.1, $CaCl_2$ 2.5 $mmol \cdot L^{-1}$, and edetic acid 1 $\mu mol \cdot L^{-1}$, pH 7.4 at 30 ℃, bubbled with 95 % O_2 + 5 % CO_2. The force of contraction was measured with a force transducer connected with a double-pen recorder (XWT-204 TYPE). Each muscle was stretched to the length at which force of contraction was maximal. The

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resting force (about 0.5 g) was kept constant throughout the experiment. Stimulation frequency was 1 Hz (the intensity was 10 % - 20 % above threshold). All preparations were allowed to equilibrate for at least 1 h until complete mechanical stabilization was obtained before any drug addition.

In the presence of β-blocker Pro 10 μmol · L^{-1}, phenylephrine (Phe) was added cumulatively in the range of 1 - 100 μmol·L^{-1}. Tissue was exposed to each concentration for 10 min.

To examine the functional changes of subtypes of α_1-adrenoceptor, the preparations were incubated with an irreversible α_{1B} antagonist chloroethylclonidine dihydrochloride (CEC) 30 μmol·L^{-1} for 30 min followed by washout before exposed to Phe.

The inotropic and chronotropic effect mediated by α_{1B} was also examined. 5-Methylurapidil (5-MU) 0.1 μmol·L^{-1} was used to block α_{1A}-adrenoceptors before Phe was added.

Spontaneously beat rate The resting force of the right heart atrium isolated from Pro- and NaCl-treated rats was adjusted to 0.3 g. Phe was added in the presence of Pro 1 μmol·L^{-1} and 5-MU 0.1 μmol·L^{-1}. The preparation was washed at least 5 times in 1 h, then carbachol (Car) was added to reduce the beat rate by 50 %, Phe was added again in the presence of β- and α_{1A}-blockers[5].

Drug used *dl*-Propranolol, phenylephrine, and carbachol (Sigma). Chloroethylclonidine dihydrochloride (Research Biochemical Inc). 5-Methylurapidil was made by Byk Gulden (Konstanz, FR Germany).

Statistical analysis Data were given as $\bar{x} \pm s$ and compared by paired t-test.

RESULTS

α_1-Adrenoceptor-mediated positive inotropic effect The basal contractility in Pro-treated rats (90 ± 18 mg) was higher than that in control group (53 ± 17 mg), $P<0.05$. In the presence of Pro, Phe produced a positive inotropic effect by stimulating α_1-aderenoceptors on papillary muscles isolated from Pro-treated rats. The effects of Phe were concentration-dependent in the range of 1 - 100 μmol · L^{-1}. Phe induced maximal increment on force of contraction was 20 ± 12 mg (n = 8) in Pro-treated rats and 5 ± 5 mg (n = 11) in NaCl-treated rats. After incubation with CEC 30 μmol·L^{-1} for 30 min, Phe produced a maximal increment on force of contraction mediated by α_{1A}-subtypes was 4 ± 4 mg (n = 5) in Pro group, which was almost the same in the control group (4 ± 5 mg, n = 7). In the presence of Pro and 5 - MU 0.1 μmol · L^{-1}, Phe induced a maximal increment on force of contraction was 13 ± 5 mg (n = 3) in Pro-treated rats. However, Phe did not induce the positive inotropic effect in NaCl-treated rats (n = 5) (Fig 1).

Spontaneous beat rate The positive chronotropic effect mediated by α_{1B} subtypes was examined on right atrium in the presence of Pro and 5-MU 0.1 μmol · L^{-1}. Phe 100 μmol · L^{-1} caused a maximal increases of 56 ± 16 beats·min^{-1} (n = 7) in Pro-treated rats and 41 ± 7 beats·min^{-1} (n = 6) in the control group.

After addition of Car for approximately 10 min, when spontaneous beating rate was reduced about 50 % and stable, the maximal increase in beat rate induced by Phe was 112 ± 18 beats·min^{-1} (n = 6) in the Pro-treated group. Similar results were found in NaCl-treated rats (107 ± 20 beats ·min^{-1}, n = 6) (Fig 2).

DISCUSSION

This study indicated that the positive inotropic effect mediated by myocardial α_1-adrenoceptors was enhanced after pretreated with Pro. While the previous studies have demonstrated that after chronic β-adrenoceptors blockade, α_1-adrenoceptor density increases in rat heart[2,3], which would be responsible for our result.

It has been known that α_1-adrenoceptor can be divided into three subtypes, α_{1A}, α_{1B}, and α_{1D} by receptor binding studies and/or molecular biological studies. However, according to their pharmacology properties, they can be mainly divided into α_{1A} and α_{1B} two groups in functional study. 5-MU and CEC are relatively selective antagonist for α_{1A} and α_{1B}, respectively[6-8]. In this study, we investigated α_{1A} and α_{1B} mediated changes in cardiac function by using 5-MU and CEC.

After preincubated with CEC, Phe induced increment on force of contraction was no significant difference between Pro- and NaCl-treated rats. As CEC was an irreversible α_{1B} antagonist, the results suggested that the positive inotropic effect mediated by α_{1A} did not change after chronic treatment with Pro. On the other hand, when in presence of 5-

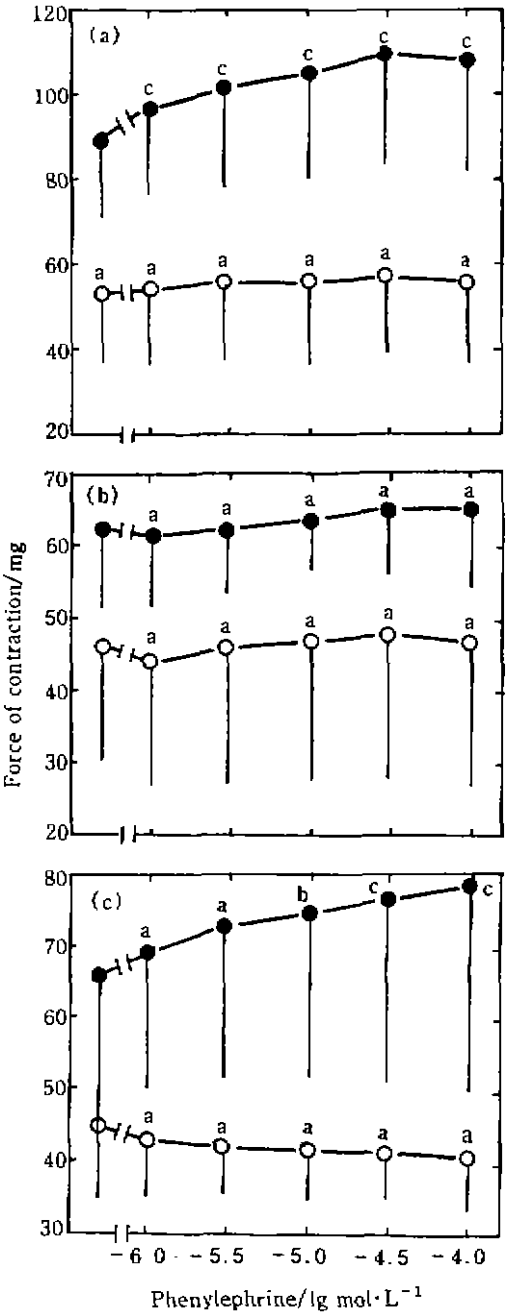

Fig 1. Effects of Phe on contraction force of left ventricular papillary muscles isolated from NaCl-(○) and Pro-(●) treated rats (a); after preincubation with CEC 30 μmol·L^{-1} (b); in the presence of 5-MU 0.1 μmol·L^{-1}(c). $n = 3-11$ rats. $\bar{x} \pm s$. $^{a}P > 0.05$, $^{b}P < 0.05$, $^{c}P < 0.01$ *vs* predrug (before Phe added) value.

MU, Phe enhanced the force of contraction significantly in Pro-treated rats, whereas, in control rats, the force of contraction were slightly decrease.

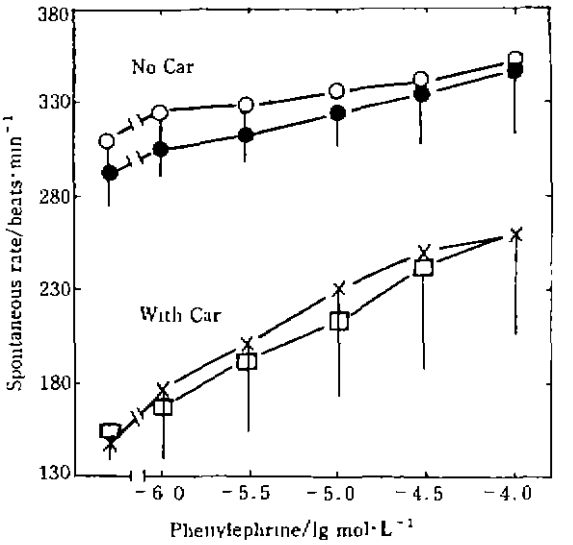

Fig 2. Effects of Phe on spontaneous beat rate in the presence of 5-MU in right heart atrium isolated from NaCl-(○), Pro-(●) treated rats without or with Car (× Pro-treated rats and □ control group). $n = 6-7$ rats. $\bar{x} \pm s$.

It may be considered that under normal condition the density of α_{1B}-subtype in the heart was more higher than that of α_{1A}-subtype. Previous study in our group showed that the density of α_{1A}-subtype increased from 19 % ± 6 % to 31 % ± 8 %, but absolutely proportion of α_{1A}-subtype was still lower than α_{1B} after β-adrenoceptors blockade[4]. The study demonstrated that α_{1B} subtype had a more important role in mediating positive inotropic effect after chronic β-adrenoceptor blockade.

The present study also showed that α_{1B} mediated positive chronotropic responses was not changed in normal physiological condition and in depressed cardiac condition after chronic β-adrenoceptor blockade.

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大鼠处理普奈洛尔后改变 α_1 受体亚型介导的心功能

李 婉，张 缨，王晓良 （中国医学科学院中国协和医科大学药物研究所，北京 100050，中国）

关键词 乳头状肌；心房；心肌收缩；心率；α_1 肾上腺素受体；普奈洛尔；苯肾上腺素；可乐定；乌拉地尔；卡巴胆碱

目的：研究普奈洛尔(Pro)作用后，大鼠心肌 α_{1A} 和 α_{1B}受体亚型介导正性肌力和正性频率变化. 方法：测定 Pro 大鼠和正常鼠左心室乳头状肌和右心房收缩力和心率. 结果：给予 Pro 后，苯肾上腺素(Phe)使乳头状肌收缩力由 53±17 mg 增加到 90±18 mg ($P<0.05$). Pro 和对照组收缩力分别增加 20±12 和 5±5 mg ($P<0.05$). 氯乙基可乐定使两组收缩力变化无区别. 5-甲基乌拉地尔存在时 Phe 使 Pro 组收缩力增加 13±5 mg，对照组无变化. 正常和心率抑制时，Phe 使两组动物 α_{1B}介导心率增加无差别. 结论：β 受体阻断，α_1 介导正性肌力增加主要由 α_{1B}作用增强引起.

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Inhibition of 11β-hydroxysteroid dehydrogenase obtained from guinea pig kidney by some bioflavonoids and triterpenoids[1]

ZHANG Yin-Di[2] (*Departments of Pharmacology and Medicine, Cornell University Medical College, 1300 York Avenue, New York NY 10021, USA*); WANG Ming-Shi (*Department of Phytochemistry, China Pharmaceutical University, Nanjing* 210009, *China*)

KEY WORDS hydroxysteroid dehydrogenases; kidney; microsomes; naringenin; emodin; fisetin; astramembranin I; oleanolic acid

AIM: To study the inhibitory effect of some bioflavonoids and triterpenoids on 11β-hydroxysteroid dehydrogenase (11β-OHSD) from guinea pig kidney. **METHOD**: The 11β-OHSD of kidney cortex microsomes in addition of cortisol was incubated in the presence of NADP, Triton DF-18, and the test compounds at 37 ℃ for 1 h. The enzyme activity was assayed by measuring the rate of conversion of cortisol to cortisone eluted with HPLC gradient analysis. **RESULTS**: The IC_{50} (95 % confidence limits)values of glycyrrhizic acid, naringenin, fisetin, emodin were 254 (202 - 320), 336 (270 - 418), 470 (392 - 564), and 527 (425 - 653) $\mu mol \cdot L^{-1}$, respectively. The inhibitory effect of oleanolic acid was 2-fold stronger than that of astramembranin I. The mode of action of naringenin was competitive inhibition. **CONCLUSION**: The test compounds inhibited the 11β-OHSD in kidney cortex with different potencies as glycyrrhizic acid did.

The syndrome of apparent mineralocorticoid excess, first described by Ulick *et al* in 1977, has led to much research on the enzyme 11β-hydroxysteroid dehydrogenase (11β-OHSD). Deficiency of 11β-OHSD in children leading to their

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[2] Now in *The Institute of Clinical Pharmacology, Nanjing Medical University, Nanjing 210029, China*.
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