Direct Action of Methoxamine on the Isolated Heart Muscle

NAOFUMI IWATSUKI

Department of Anesthesiology,* Tohoku University School of Medicine, Sendai

IWATSUKI, N. Direct Action of Methoxamine on the Isolated Heart Muscle. Tohoku J. exp. Med., 1972, 108 (3), 245–252 — A direct effect of methoxamine on the contractile state of heart muscle was determined by examining its effects on isometric peak force (F), maximum velocity of force development (dF/dt) and time to peak force (TPF) in isolated canine trabeculae. Methoxamine at the concentrations over $5 \times 10^{-5}$M caused a concentration-dependent depression of F and dF/dt. TPF was slightly shortened at higher concentrations of methoxamine (over $1 \times 10^{-4}$M). These results suggest that methoxamine possesses a direct negative inotropic action which is mainly due to a decrease in the intensity of the active state. An increase in F produced by isoproterenol was antagonized by methoxamine at a concentration about 100 times higher than that of isoproterenol, suggesting a weak beta-adrenergic blocking action of methoxamine. This antagonistic action of methoxamine was seen also in the effects of isoproterenol on dF/dt as well as TPF. The effect of methoxamine on the mechanics of muscle contraction is qualitatively similar to, but quantitatively different from that of other beta-adrenergic blockers. — methoxamine; isolated heart muscle; inotropism; beta-adrenergic blocker

Early studies suggested that methoxamine, a pure alpha-adrenergic stimulator, might exert some direct effects on the heart (Melville and Lu 1952, Goldberg et al. 1960, Imai et al. 1961, Smith and Whitcher 1967). Recently it was demonstrated in isolated heart muscle preparations that methoxamine caused a decrease in tension at isometric contraction, suggesting a direct negative inotropic action of this drug (Blinks 1964). However, tension depends on two factors at isometric contraction: velocity of tension development and duration during which the tension is generated. Velocity of tension development depends on velocity of shortening of the contractile element under unchanged stiffness of the elastic element (Hill 1938). Duration of tension development is reflected on the duration of chemical processes which take place during muscle contraction (Sonnenblick 1967). Therefore, measuring velocity and duration of tension development seems to be more pertinent to assessing the changes in intrinsic contractile properties of the heart muscle than measuring tension alone.

The present study was undertaken to evaluate the changes in intrinsic contractile properties of the heart muscle produced by methoxamine by measuring

Received for publication, April 3, 1972.

* Director: Prof. K. Iwatsuki.
velocity and duration of isometric force development in isolated canine trabeculae. An interaction of methoxamine with isoproterenol, a beta-adrenergic stimulator, on the heart muscle was also investigated in this study.

MATERIALS AND METHODS

Trabeculae were excised from the right ventricle of healthy mongrel dogs anesthetized with pentobarbital sodium (25 mg/kg i.v.). The muscles were suspended vertically in a muscle bath surrounded by a thermo-bath with circulating water which was kept at 27°C by Coolnics Thermo-Bath (CTE-2). The muscle bath was filled with 180 ml of Krebs-Henseleit solution, bubbled with a 95 per cent oxygen and 5 per cent carbon dioxide gas mixture. The composition of Krebs-Henseleit solution in mmole per liter was as follows: NaCl 118, KCl 4.7, CaCl₂ 2.5, MgSO₄ 1.2, NaHCO₃ 21, KH₂PO₄ 1.2 and glucose 5.6. Each end of the trabecula was held by a springed clip to which a steel wire was attached. The lower end of the trabecula was connected to the transducer (Nihon Kohden SB-1T) and the upper end was connected to a longer side of the lever system through the steel wires. A shorter side of the lever system was fixed by a micrometer which was adjusted according to the length of muscle. Electrical stimulation was provided through platinum electrodes (0.5 cm × 2.0 cm) placed along the entire length of the muscle. A square wave of 5 msec duration with a voltage less than 20 per cent above threshold was applied at a frequency of 18 per minute by the stimulator (Nihon Koden MSE-3) (Fig. 1). The muscle was allowed to contract more than one hour after dissection for the stabilization of contraction. Then the muscle was fixed, using a micrometer, at a length at which maximum isometric force was obtained (Lmax). The length of Lmax was measured with the telescope (Pika PRM-2) placed in front of the muscle chamber. Each measurement was made at Lmax and at a steady state of contraction. The drugs for the study were added directly into the bathing solution. The solution was replaced completely by a fresh solution five to six times after each series of measurement. Isometric
force and velocity of force development which was obtained by means of an R-C differentiating circuit (time constant: 0.6 msec) were recorded with a marker of electrical stimulation on direct-writing papers at a paper speed of 60 mm per second. All values were expressed as mean±SE and analyzed statistically by Fisher’s t-test for paired data.

RESULTS

Concentration–Response Relationship

Ten measurements were done in each of 6 preparations. Muscle length, blotted weight and the cross-sectional area of 6 trabeculae were 7.50±0.36 mm, 16.6±3.1 mg and 2.17±0.31 mm², respectively. The concentrations of methoxamine used were $2 \times 10^{-5}$M, $5 \times 10^{-5}$M, $1 \times 10^{-4}$ and $2 \times 10^{-4}$M.

Methoxamine at a concentration of $2 \times 10^{-5}$M produced no significant changes either in isometric peak force (F) or maximum velocity of force development (dF/dt). However, when the concentration of methoxamine was increased over $5 \times 10^{-5}$M, methoxamine caused a concentration-dependent depression of both F and dF/dt (Figs. 2 and 3). At the concentrations of $5 \times 10^{-5}$M, $1 \times 10^{-4}$M and $2 \times 10^{-4}$M, the mean per cent values of F were 96.1±0.7, 91.5±1.0 and 82.7±1.7 of the control and those of dF/dt were 96.7±0.6, 91.3±1.4 and 83.3±1.9 of the control, respectively ($P$ values for each, <0.001).

Methoxamine at the concentrations of $2 \times 10^{-5}$M and $5 \times 10^{-5}$M did not change the time from the onset of isometric contraction to isometric peak force (TPF), but at the concentrations of $1 \times 10^{-4}$M and $2 \times 10^{-4}$M, it caused decreases in TPF to 96.1±1.2% and 95.2±1.6% of the control, respectively ($P$ values for each, <0.02) (Fig. 4). Methoxamine tended to prolong the time from the stimulation to the onset of isometric contraction (TOF) following an increase in its concentration, but these changes were not statistically significant (Fig. 4).

![Fig. 2. Dose-response relationships between methoxamine and isometric peak force (F). Abscissa: concentrations of methoxamine on log scale. Ordinate: mean per cent values of F to the control. Note: dose-dependent decreases in F.](image-url)
Fig. 3. Dose-response relationships between methoxamine and maximum velocity of force development (dF/dt). Abscissa: concentrations of methoxamine in M on log. scale. Ordinate: mean per cent values of dF/dt to the control. Note: the slope of dose-dependent decreases in dF/dt is close to that in F.

Fig. 4. Mean per cent values of the time to peak force from the onset of force development (TPF) and the time to onset of force development from the stimulation (TOF) are plotted against methoxamine concentrations in M on log scale.

Interaction of Methoxamine with Isoproterenol

The concentrations of isoproterenol used were $2.2 \times 10^{-6}$M, $2.2 \times 10^{-7}$M, and $2.2 \times 10^{-8}$M. Four preparations were used in each study. Muscle length, blotted weight and the cross-sectional area were $7.14 \pm 0.34$ mm, $14.8 \pm 2.6$ mg and $2.06 \pm 0.3$ mm$^2$ at the study of $2.2 \times 10^{-6}$M of isoproterenol, $5.88 \pm 0.47$ mm, $8.38 \pm 1.91$
mg and 1.38±0.24 mm² at the study of 2.2×10⁻⁷M and 6.08±0.46 mm², 10.0±2.06 mg and 1.60±0.28 mm² at the study of 2.2×10⁻⁸M, respectively.

The administration of 1×10⁻⁴M and 2×10⁻⁴M of methoxamine did not cause any decrease in F under the influence of 2.2×10⁻⁶M of isoproterenol (Fig. 5). The effect of 2.2×10⁻⁷M of isoproterenol on F was not significantly affected by adding 2×10⁻⁴M of methoxamine (P>0.05), but its effect on dF/dt was inhibited (P<0.02). However, the effects of 2.2×10⁻⁸M of isoproterenol on F and dF/dt were both inhibited by adding 2×10⁻⁴M of methoxamine (P<0.05 and <0.02, respectively). Consequently, the concentration-response curves of isoproterenol for F and dF/dt were shifted to the left by methoxamine (Fig. 6).

**DISCUSSION**

The present study showed that methoxamine caused a dose-dependent decrease in isometric peak force (F) of the isolated heart muscle. Such an effect was seen at the concentrations more than 5×10⁻⁴M of methoxamine. The results are consistent with those of the previous study done using a strip of the left atrium (Blinks 1964).

Isometric peak force depends on two factors: velocity of force development and duration during which the force is generated. Since the values of per cent decrease in F at each concentration of methoxamine were close to those in dF/dt, a decrease in F produced by methoxamine seems to be due mainly to a decrease in velocity of force development. The influence of a decrease in duration of force.
Fig. 6. The influence of methoxamine to the effects of isoproterenol on isometric peak force (F) and maximum velocity of force development (dF/dt) are expressed by plotting the per cent changes in F and dF/dt on ordinate against the concentrations of isoproterenol in M on abscissa. Note: both isoproterenol–F and isoproterenol–dF/dt curves are shifted leftward under the influence of methoxamine 2 × 10^{-4}M (open circles).

Development on F was small and it was seen only at higher concentrations of methoxamine, as shown by minor changes in the time from the onset of isometric contraction to peak force (TPF) in the present study.

According to Hill's model of muscle contraction, the contractile element (CE) is a fundamental property responsible for generating force and the series elastic element (SE) is a passive spring. Thus, dF/dt at isometric contraction reflects velocity of shortening of CE, when stiffness of SE is unchanged (Hill 1938). As the changes in resting tension were not observed following the administration of methoxamine, the state of SE might not be altered by methoxamine. The decrease in dF/dt, therefore, seems to be due to a direct depressive effect of methoxamine on CE. In other words, methoxamine possesses a direct negative inotropic action. Sonnenblick (1967) demonstrated that dF/dt is closely related to the intensity of the active state* and TPF is directly proportional to the duration of the active state. The results of the present study, therefore, suggest that a negative inotropic action of methoxamine is mainly due to a decrease in the intensity of the active state and in a small part due to a shortening of the duration of the active state.

A leftward shift of the concentration–response curve of isoproterenol for F and dF/dt by methoxamine suggests that methoxamine possesses a beta-adrenergic

* The active state is a mechanical measure, in terms of force and velocity of shortening, of chemical processes which take place during muscle contraction.
receptor blocking action. However, its potency as a beta-adrenergic blocker is weak, since the ratio of minimum concentration of methoxamine as an antagonist to isoproterenol was about 100 to 1 on molar basis in the present study, whereas the ratio of pronethalol to norepinephrine was reported to be about one to one (Koch-Weser 1964). Moreover, a relative potency of pronethalol as a beta-adrenergic blocker was one-seventh of that of propranolol (Levy and Richards 1965). These results are consistent with those of the previous studies (Imai et al. 1961, Blinks 1964). An interesting finding of the present study was that an antagonism of methoxamine to the effect of isoproterenol on dF/dt observed at higher concentrations of isoproterenol than that on F (Fig. 6). This result may be explained by prolonged TPF produced by methoxamine, consequently resulting in a smaller decrease in F than in dF/dt. The decrease in dF/dt and the prolongation of TPF produced by methoxamine under the presence of isoproterenol in the present study suggest that methoxamine antagonizes the both effects of isoproterenol on dF/dt and TPF.

It should be pointed out that a major decrease in dF/dt associated with a minor decrease in TPF produced by methoxamine is in a similar manner to that observed following the administration of beta-adrenergic blockers such as pronethalol (Koch-Weser 1964) and propranolol (Buccino et al. 1967). The effect of methoxamine on the mechanics of muscle contraction is, therefore, qualitatively similar to, but quantitatively different from that of other beta-adrenergic blockers.

Prolongation of the time from the stimulation to the onset of contraction (TOF) produced by methoxamine in the present study suggests that methoxamine may affect the time from excitation to contraction of the muscle. This result may be related to an anti-arrhythmic action of this drug (Lahti et al. 1955).

A clinical dose of methoxamine ($2 \times 10^{-5}$M) did not show any depression on the contractile state of the isolated heart muscle in the present study. From this result it may be assumed that methoxamine within clinical doses does not exert any harmful effects on the normal myocardium. However, a possibility cannot completely be denied that it may act as a depressant to the impaired myocardium even in a clinical dose. A depressive action of methoxamine to the myocardium can easily be counteracted by beta-adrenergic stimulators.

CONCLUSION

Methoxamine exerts a direct negative inotropic effect on the heart muscle. This effect is mainly due to a decrease in the intensity of the active state.

Methoxamine possesses a weak beta-adrenergic blocking action. The effect of methoxamine on the mechanics of muscle contraction is qualitatively similar to, but quantitatively different from that of other beta-adrenergic blockers.

References


