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Electrophysiologic and Inotropic Actions of Prostaglandin $F_{2\alpha}$ in Rat Myocardium (38351)

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Several prostaglandins have been shown to elicit positive inotropic responses in myocardium from several species (1–4) although species specificity is recognized. Reports also have appeared describing prostaglandin E_1 induced alterations of the cardiac action potential. In frog myocardium, prostaglandin E_1 is reported to cause an increase in the amplitude and duration of the action potential (5), while in guinea pig hearts a decrease in the action potential duration and an increase in the resting potential are found (6).

This report describes electrophysiologic effects of prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$) accompanying the inotropic response in isolated rat papillary preparations. $PGF_{2\alpha}$ has been shown to elicit a strong positive inotropic response in intact rat hearts (7).

Methods. Left ventricular posterior papillary muscles were rapidly excised from hearts of 200–300 g Sprague–Dawley white male rats under ether anesthesia. The muscle base along with some attached ventricle was gripped in a lucite clamp and mounted in a 5 ml capacity horizontal muscle bath. Muscles were bathed at 37° with oxygenated Krebs–Ringer's solution (8) of pH 7.3 and containing 2.5 m*M* Ca^{2+}. The preparations were driven electrically at 12 stimuli per min with 0.5 msec suprathreshold square wave pulses and twitch tension was measured using a Statham 0.3 oz isometric tension transducer. Transmembrane potentials were measured with 3 *M* KCl filled glass microelectrodes having 10–30 megohms resistance and connected through calomel half cells to a Medistor A35 electrometer. Tension and transmembrane potential data, along with the first time derivative of each, were recorded simultaneously using four channels of a Honeywell Visicorder with galvanometers having a frequency response linear to 2×10^3 Hz.

All muscles were allowed to equilibrate for one hour during which the preload yielding the peak tension in the length–tension relationship was determined. Thereafter, this preload was maintained throughout the experiment. Following equilibration, control data were collected over a ten minute period. Subsequently, the chamber was drained and perfused with fresh Krebs–Ringer's solution containing $PGF_{2\alpha}$. Fifteen minutes later, with the full inotropic effect present, the ten minute period of test data collection was initiated. $PGF_{2\alpha}$ in crystalline form was initially solubilized in 95% ethyl alcohol (5 mg/ml) from which working stocks were prepared as needed. The concentrations studied were 10 ng/ml and 100 ng/ml. Data were analyzed by the small sample *t* test with each muscle serving as its own control.

Results. One group of muscles ($N = 10$), to which no $PGF_{2\alpha}$ was applied during the test period, was studied to ascertain the stability of the rat papillary preparation with time. For all parameters measured, the performance of the preparation remained unchanged ($P > 0.05$) throughout the control and test periods. Two additional muscles were studied to detect the possible effect of ethyl alcohol in the test solution at a concentration equivalent to that encountered with the highest $PGF_{2\alpha}$ concentration used. At this level, ethyl alcohol produced no effect ($P > 0.05$) on the electrical or mechanical responses. One group of muscles ($N = 10$) was exposed to 100 ng/ml $PGF_{2\alpha}$ during the test period; another group of muscles ($N = 4$) was exposed to 10 ng/ml $PGF_{2\alpha}$ to test for dose dependency. With $PGF_{2\alpha}$ perfusion, a dose related increase in the peak twitch tension was always

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observed. Time to peak tension was not significantly altered. Dose related increases in the maximum rate of tension development and the maximum rate of relaxation were found.

PGF$_{2\alpha}$, in either concentration tested, was found to have no significant effect on the membrane resting potential. Similarly, no significant effect was found on the action potential spike amplitude. However, a dose related decrease in the maximum rate of depolarization was observed. The action potential duration measured at the 0 mV level of polarization, the −60 mV level of polarization, and at a level of polarization corresponding to 50% of the action potential spike amplitude, was prolonged, sometimes by up to 60%.

Tracings of a control record and a 100 ng/ml PGF$_{2\alpha}$ record from the same muscle are shown in

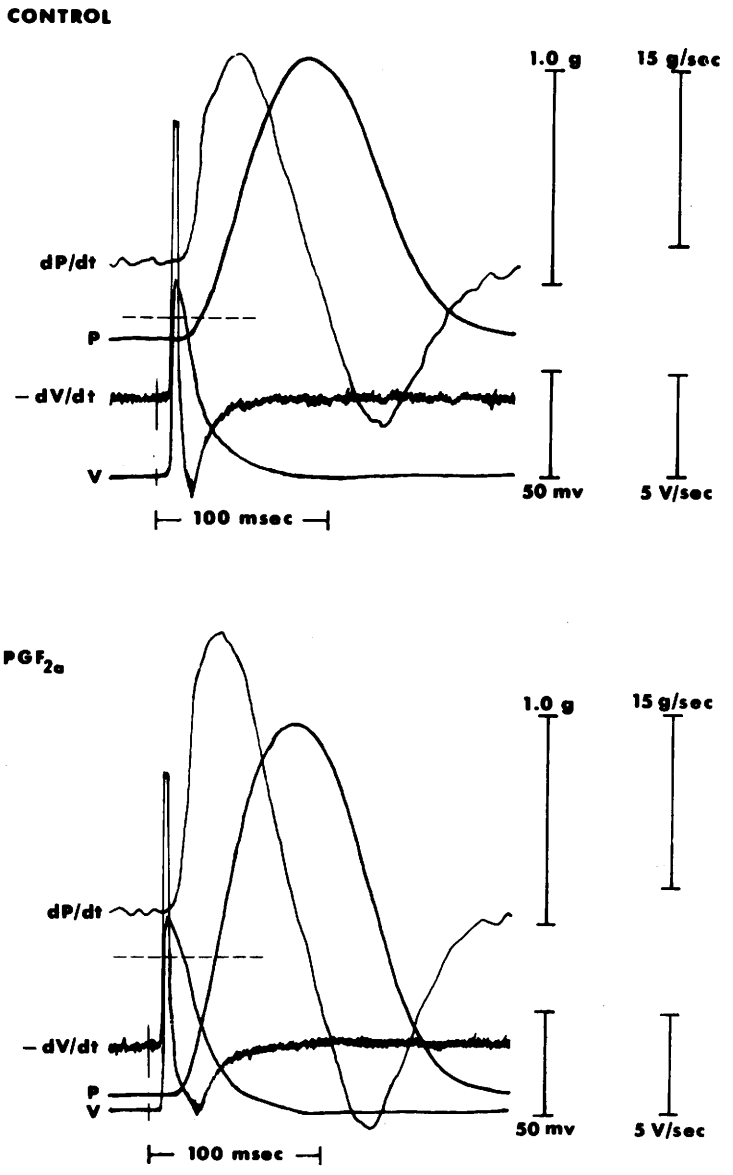

FIG. 1. Tracings of a control record and a 100 ng/ml PGF$_{2\alpha}$ record from the same muscle. P is isometric twitch tension, dP/dt is the rate of tension change, V is the membrane potential, −dV/dt is the rate of repolarization. Respective calibrations are given by the vertical lines to the right of the figure. The horizontal dashed line in each record represents the isopotential line.

Fig. 1. Differences to be noted between the control and prostaglandin records include: an increase in the twitch tension (P), an increase in the maximum rate of tension development (dP/dt), an increase in the action potential (V) duration and plateau amplitude, and a decrease in the maximum rate of repolarization ($-dV/dt$). Changes in the rate of depolarization are not shown. Data for all muscles are summarized in Table I.

Discussion. Our results demonstrate that $PGF_{2\alpha}$ exerts consistent electrophysiologic as well as positive inotropic effects in isolated rat papillary muscle. Electrophysiologic alterations with $PGF_{2\alpha}$ have not been previously reported, and were found to involve mainly the plateau and repolarization phases of the action potential. $PGF_{2\alpha}$ caused an increase in the action potential duration and plateau amplitude. A modest decrease in the maximum rate of depolarization also was observed. The peak twitch tension was augmented, the increase due to an increased rate of tension development while time to peak tension remained unchanged. The effects of $PGF_{2\alpha}$ seemed more pronounced at the higher concentration tested, suggesting a dose dependency.

These results are consistent with those of Smejkal *et al.* (5). Using a sucrose gap technique to measure potential, they found that PGE_1 (10^{-7}–10^{-5} M) increased the amplitude and duration of frog atrial action potentials. PGE_1 also is known to exert positive inotropic effects in frog hearts (1). In Langendorff perfused guinea pig hearts, however, PGE_1 (5 μg bolus) is reported to reduce slightly the action potential duration, increase the resting potential, and produce a positive inotropic effect (6).

In cardiac muscle preparations from several species, two inward going membrane current components have been identified; a rapid inward current carried by Na^+ ions, and a slow inward current carried by Ca^{2+}, or by Ca^{2+} and Na^+ ions (9–11). These inward current components have also been demonstrated for rat ventricular fibers (12). The rapid inward current is considered mainly responsible for the action potential upstroke, with the slow inward current contributing chiefly to maintenance of the action potential plateau. Repolarization of the cardiac action potential then ensues with inactivation of the slow inward current, activation of outward current, or both in combination (13,14). In addition to electrophysiologic significance, the slow inward current also has important implications in cardiac muscle contraction (11, 15, 16).

The results of our experiments could be explained simply by hypothesizing that $PGF_{2\alpha}$ induced an augmentation of the slow inward current. Such augmentation of the slow inward current, due to increased inward movement of Ca^{2+},

TABLE I. Effects of $PGF_{2\alpha}$ on Electrical and Mechanical Parameters of Rat Papillary Muscle.

	10 ng/ml		100 ng/ml	
Parameter[a]	Control	Experimental	Control	Experimental
APD (msec) 0 mV	7.0 ± 0.5	7.8 ± 0.7	5.0 ± 0.3	7.8 ± 0.5[b]
−60 mV	48.5 ± 2.4	58.8 ± 3.9[b]	46.2 ± 4.1	64.5 ± 3.6[b]
50% AP Amp	14.2 ± 0.5	18.0 ± 0.9[b]	10.2 ± 0.5	15.1 ± 0.7[b]
dV/dt (V/sec)	88.7 ± 2.3	81.5 ± 2.0[b]	90.8 ± 2.4	74.4 ± 4.2[b]
−dV/dt (V/sec)	4.7 ± 0.2	3.8 ± 0.2[b]	5.4 ± 0.3	3.6 ± 0.2[b]
RP (mV)	69.2 ± 0.6	70.7 ± 0.8	69.6 ± 0.6	68.9 ± 0.8
AP (mV)	86.9 ± 1.4	88.0 ± 1.2	85.8 ± 1.0	88.1 ± 1.2
P (mg/mm²)	945 ± 35	1064 ± 35[b]	1055 ± 49	1331 ± 57[b]
TPT (msec)	74.7 ± 1.3	74.8 ± 1.3	73.6 ± 0.6	74.9 ± 0.8
dP/dt (g/sec)	20.13 ± 0.87	23.49 ± 0.86[b]	20.80 ± 1.06	27.41 ± 1.44[b]
−dP/dt (g/sec)	15.16 ± 0.86	18.42 ± 0.88[b]	17.34 ± 0.83	21.56 ± 0.91[b]

[a] Data ($\bar{X}$ ± SE) expressed are for both $PGF_{2\alpha}$ concentrations tested and the respective control groups. ADP is action potential duration measured at the 0 mV level, the −60 mV level, and 50% of the action potential amplitude; dV/dt and $-dV/dt$ are maximum rates of depolarization and repolarization, respectively; RP is resting potential; AP is action potential amplitude; P is maximum twitch tension; TPT is time to peak tension; dP/dt and $-dP/dt$ are maximum rates of tension development and relaxation, respectively.

[b] $P < 0.05$ compared to respective control.

or Ca^{2+} and Na^{+} ions, would be expected to cause a more depolarized and longer lasting action potential plateau and also could, at least in part, account for the positive inotropic response. A similar hypothesis has been suggested for the cardiac action of epinephrine, which in addition to its well known inotropic influence, causes an increase in the action potential plateau and in the magnitude of the slow inward current (17, 18).

Alternative hypotheses which include a $PGF_{2\alpha}$ influence on the rapid inward current, as suggested by the observed decrease in the maximum rate of depolarization (Table I), and on the outward current cannot be excluded. Voltage clamp studies of the specific membrane currents would be of value in determining the validity of the various hypotheses.

Summary. The effects of $PGF_{2\alpha}$ on electrophysiologic and mechanical parameters in electrically driven rat papillary muscles were studied. $PGF_{2\alpha}$ (100 ng/ml) was found to reduce slightly the maximum rate of rise of the action potential while increasing the plateau amplitude and duration. The magnitude of the twitch and the maximum rate of tension development were augmented whereas time to peak tension was unchanged. At a lower concentration, $PGF_{2\alpha}$ (10 ng/ml) produced similar results, but of reduced magnitude.

The results may be explained by hypothesizing that $PGF_{2\alpha}$ acts upon ionic conductances possibly by augmenting the slow inward current in rat myocardium.

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1. Berti, F., Lentati, R., and Usardi, M. M., Med. Exp. **13,** 233 (1965).

2. Vergroesen, A. J., DeBoer, J., and Gottenbos, J. J., *in* "Nobel Symposium II: Prostaglandins" (S. Bergstrom and B. Samuelson, ed.), p. 211. Interscience Publishers, New York (1967).

3. Levy, J. V., and Killebrew, E., Proc. Soc. Exp. Biol. Med. **136,** 1227 (1971).

4. Nutter, D. O., and Ratts, T., Prostaglandins **3,** 323 (1973).

5. Smejkal, V., Rougier, O., and Garnier, D., Physiol. Bohemoslov. **19,** 23 (1970).

6. Koboyaski, T., Nakayama, R., and Kimura, K., Jap. Circ. J. **35,** 807 (1971).

7. Vergroesen, A. J., DeBoer, J. and Gottenbos, J. J., Acta Physiol. Pharmacol. Neerl. **15,** 72 (1969).

8. Henderson, A. H., Brutsaert, D. L., Parmley, W. W., and Sonnenblick, E. H., Amer. J. Physiol. **217,** 1273 (1969).

9. Rougier, O., Vassort, G., Garnier, D., Gargouil, Y. M., and Coraboeuf, E., Pfluegers Arch. **308,** 91 (1969).

10. Beeler, G. W., and Reuter, H., J. Physiol. (London) **207,** 191 (1970).

11. New, W., and Trautwein, W., Pfluegers Arch. **334** 24 (1972).

12. Besseau, A., and Gargouil, Y. M., J. Physiol. (London) **204,** 95P (1969).

13. Nobel, D., and Tsien, R. W., *in* "Electrical Phenomena in the Heart" (W. C. DeMello, ed.), p. 133. Academic Press, New York (1972).

14. Trautwein, W., Physiol. Rev. **53,** 793 (1973).

15. Beeler, G. W., and Reuter, H., J. Physiol. (London) **207,** 211 (1970).

16. Vassort, G., and Rougier, O., Pfluegers Arch. **331,** 191 (1972).

17. Reuter, H., Pfluegers Arch. **287,** 357 (1966).

18. Vassort, G., Rougier, O., Garnier, D., Sauviat, M. P., Coraboeuf, E., and Gargouil, Y. M., Pfluegers Arch. **309,** 70 (1969).

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