EFFECTS OF ADRENALINE AND NORADRENALINE ON THE CARDIOVASCULAR SYSTEM

REPORT 4: EFFECT OF STIMULUS FREQUENCY ON PAPILLARY CONTRACTION*

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INTRODUCTION

A few investigators (1-3) have reported the negative inotropic action of adrenaline on isolated mammalian hearts. In the previous report (4), we have demonstrated that this effect in fresh isolated heart is of sympathetic origin. However, as yet, detailed investigations on the effect and its mechanism have not been reported. It is supposed that condition of the fresh isolated preparation is more closer to those of the living animal than the elapsed preparation. Hence, adrenaline negative inotropic effect of the fresh isolated heart should likely to concern in the mechanism of the cardiac inhibition by adrenaline in living animals. Y. Kobayashi (5) has reported that the Langendorff's preparation with the heart rate of living animals were usually sensitive to the various drugs and suitable for bioassay.

Since the adrenaline negative inotropism can occur simultaneously with positive chronotropism only with the fresh preparation whose heart rate kept above approximately 200 per minute, it was presumed, then, adrenaline positive chronotropism did imparted a reasonable effect on the inotropic action. Therefore, the authors have attempted to investigate the influence of increased electric stimulus frequency upon the amplitude of contraction of rabbit papillary muscle.

PROCEDURE

White male rabbits weighing approximately 2.5 kg were used. The experiment was performed by the method previously described by McK. Cattell and H. Gold (6). The papillary muscle was removed from the right ventricle and was stimulated electrically at approximately 0.6 V by a thyatron which was designed by T. Fukuhara (7). The stimulus frequency was changed between 30 to 320 per minute. The papillary contractions were recorded by optical method. A mixture of defibrinated blood and Locke's solution

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at the volume ratio of 1:20 was employed as the papillary bath solution and throughout the experiment the solution was kept at a temperature of 35°C and was saturated with oxygen. *L*-adrenaline hydrochloride (Daiichi Seiyaku Co. Ltd.) and *DL*-noradrenaline hydrochloride (Sankyo Co. Ltd.) were used.

RESULTS

1) Effect of increased frequency

In most cases, as in Fig. 1, no diminution was found of the amplitude of contraction with the increased frequency from 40 to 60 per minute.

While, the amplitude of contraction was usually reduced but occasionally augmented (Fig. 2) with the increase of frequency from 32 to 102 per minute.

The increase of frequency from 100 to 136 per minute induced a slight decrease of the maximum levels of contraction and simultaneously a slight elevation of relaxation levels (Fig. 3). In general, the initial few contractions immediately following the sudden transition from low to high frequency were slightly diminished and returned gradually to those in the basic frequency. And following the abrupt returning to the basic frequency, the subsequent few beats have been augmented. These results have supported what some investigators (8, 9) have reported previously. However, in the records shown in Fig. 1 and 3, the frequency-induced potentiation was not seen. In most of present experimental series, there were some differences in the amplitude of contraction before and after the frequency change.

For example, the record in Fig. 4 shows that a marked diminution of the amplitude has occurred by the increased frequency from 150 to 265 per minute. As in the above series of experiments, the elevation of relaxation levels tend to occur along with the decrease of the maximum contraction levels.

The records obtained from some papillaries indicated, as in Fig. 5, that a remarkable elevation of relaxation levels, consequently a marked diminution of the amplitude was elicited by the increased frequency from 150 to 320 per minute. In these cases, the insufficient relaxation resulting

FIG. 1

FIG. 2

FIG. 3

FIG. 4

FIG. 5

150/min ↑ 274/min ↓ 160/min

FIG. 6

200/min ↑ 232/min ↓ 150/min

FIG. 7

150/min ↑ 293/min ↓ 150/min

FIG. 8

200/min ↑ 268/min ↓ 200/min

FIG. 9

150/min ↑ 237/min ↓ 150/min

FIG. 10

from high frequency may be responsible for this phenomenon.

And, some results, as in Fig. 6, were characterized by a remarkable elevation of relaxation levels, consequently, a diminution of the amplitude and by the initial depression immediately following the sudden increase of frequency and the post-stimulation potentiation noted by H. Rosin *et al.* (8).

In some cases, as the record presented in Fig. 7, the irregularity of the contraction was induced by the increased frequency from 200 to 232 per minute.

2) Influences of adrenaline and noradrenaline on the effect induced by the increased frequency

When large doses of *L*-adrenaline hydrochloride (final concentration; 1.5×10^{-6}) were added to the papillary bath solution, also, a marked diminution of the maximum contraction levels was observed following the increase of frequency from 150 to 293 per minute (Fig. 8) and from 200 to 268 per minute (Fig. 9).

The results obtained in similar experiments with added *DL*-noradrenaline hydrochloride (3×10^{-6}) were essentially the same as with *L*-adrenaline (Fig. 10).

Even in the experiments with the addition of these agents, the initial depression following the sudden increase of frequency and the post-stimulation potentiation were

observed. And the diminution of the maximum contraction levels appeared to be more remarkable than those in the absence of these agents.

After prolonged high frequency stimulation (240/min), the basic frequency (150/min) failed to induce recovery of the amplitude.

DISCUSSION

In regard to the relationship between the force and frequency of contraction, McK. Cattell *et al.* and others (8-20) demonstrated the augmentation with increased frequency. Some investigators have looked into the mechanism of the potentiation resulting from

rapid stimulation. S. Hajdu *et al.* (21) proposed potassium movement to explain staircase in the frog, namely, the high frequency induces the insufficiency of time for the K^+ lost during contraction to reenter the fiber and the low content of K^+ raises tension. H. Rosin *et al.* (8) and W. J. Whalen *et al.* (13, 14) attempted to explain it on the basis of a potentiating substance (probably *noradrenaline*). While, P. B. Hollander *et al.* and others (22-26) observed a decrease in amplitude of contraction when the stimulus frequency was increased. In our experiment, the contraction amplitude had decreased explaining an evidence that the adrenaline positive chronotropism had engaged the negative inotropic process partly. In extremely high frequency stimulation, the papillary contraction may be depressed because of the excessive loss of intracellular K^+ as described by S. Hajdu (27). In other words, when adrenaline had been administered to the isolated heart whose rate was equal to that of living one, the amplitude of heart beat could have been decreased even with very existence of positive chronotropic effect. It seems certain that the adrenaline negative inotropic effect could be produced by the positive chronotropic action, nevertheless, the reasons offered are far from satisfaction to elucidate fully and there are more angles to be scrutinized.

Generally, it is considered that the positive chronotropic effect of adrenaline can reduce the amplitude of cardiac contraction and the metabolic action seemed to augment the amplitude. In the fresh isolated heart whose beat is more rapid, the influence of the positive chronotropic effect of adrenaline upon the cardiac contraction may exert more potent influence than the metabolic action, on the other hand, it may be that the metabolic action may exert stronger influence than positive chronotropic action when the positive inotropic effect was elicited in the older preparation.

Since the maintenance of heart rate over 200 per minute of an isolated preparation was an important factor for the adrenaline negative inotropic effect, it is probable that the condition of rapid heart rate itself may be responsible for the chief cause of negative inotropism. It has been known that the sympathetic nerve plays an important role on the maintenance of heart rate at normal level. If so, denervation of the sympathetic nerve would likely to influence the adrenaline negative inotropic effect. The influence of sympathetic denervation will be demonstrated elsewhere. It seemed apparent that the humoral substance to be released from the sympathetic nerve endings should keep the heart rate high. Hence, the influences of sympatholytics and other autonomic blocking agents on the adrenaline negative inotropic effect are being studied.

It is author's contention that any method to produce rapid heart beat is possible to make adrenaline negative inotropic effect, which can be blocked by any cause that makes the heart beat slower.

SUMMARY

1. The contraction amplitude of the rabbit papillary muscle was not remarkably changed when the stimulus frequency was increased from 40 to 60 or from 100 to 136 per minute.

2. The amplitude of contraction was usually reduced but occasionally augmented with increase of frequency from 32 to 102 per minute.

3. The amplitude of contraction was reduced when the stimulus frequency increased as follows; from 150 to 265, from 150 to 274, from 150 to 320 and from 200 to 232 per minute.

4. When *L*-adrenaline (final contraction; 1.5×10^{-6}) or *DL*-noradrenaline (3.0×10^{-6}) was added to the papillary bath solution, also, a significant diminution of the contraction amplitude was induced by the increased frequency.

CONCLUSION

The positive chronotropic effect of adrenaline plays an important role on the negative inotropic effect produced by this substance.

REFERENCES

- 1) ANDO, J. *et al.*: *Bull. Osaka Med. School* **4**, 49 (1958)
- 2) CLAES, M.E.: *C. R. Soc. Biol., Paris* **87**, 783 (1922)
- 3) SERIN, F.: *Acta Physiol. Scand.* **26**, 291 (1952)
- 4) ANDO, J. *et al.*: *THIS JOURNAL* **9**, 14 (1959)
- 5) KOBAYASHI, Y.: *Yakurigaku Jishu Kogi* P. 239, Nankodo, Tokyo (1938)
- 6) CATTELL, MCK. AND GOLD, H.: *J. Pharmacol.* **62**, 116 (1938)
- 7) FUKUHARA, T.: *Seirigaku Jikken-ho* P. 31, Nanzando, Tokyo (1956)
- 8) ROSIN, AND FARAH, A.: *Amer. J. Physiol.* **180**, 75 (1955)
- 9) CATTELL, MCK. AND GOLD, H.: *Ibid.* **182**, 307 (1955)
- 10) CATTELL, MCK. AND GOLD, H.: *Ibid.* **133**, 236 (1941)
- 11) KRUTA, V.: *Arch. internat. Physiol.* **45**, 332 (1937)
- 12) HOFFMAN, B.F., BENDLER, E. AND SUCKLING, E.E.: *Amer. J. Physiol.* **185**, 95 (1956)
- 13) WHALEN, W.J.: *Science* **127**, 468 (1958)
- 14) WHALEN, W.J., FISHMAN, N. AND ERICKSON, R.: *Amer. J. Physiol.* **194**, 573 (1958)
- 15) ROSIN, H. AND FARAH, A.: *Fed. Proc.* **13**, 400 (1954)
- 16) LEE, K.S.: *J. Pharmacol.* **112**, 484 (1954)
- 17) HAJDU, S. AND SZENT-GYÖRGYI, A.: *Amer. J. Physiol.* **168**, 159 (1952)
- 18) HAJDU, S. AND SZENT-GYÖRGYI, A.: *Ibid.* **168**, 171 (1952)
- 19) FURCHGOTT, R.F. *et al.*: *J. Pharmacol.* **110**, 19 (1954)
- 20) SZENT-GYÖRGYI, A.: *Chemical Physiology of Contraction in Body and Heart Muscle* Acad. Press, New York (1953)
- 21) HAJDU, S.: *J. Pharmacol.* **120**, 90 (1957)
- 22) HOLLANDER, P.B. AND WEBB, J.L.: *Circulation Res.* **3**, 604 (1955)
- 23) McDOWELL, R.J.S. *et al.*: *J. Physiol.* **130**, 615 (1955)
- 24) KATZUNG, B. AND FARAH, A.: *Amer. J. Physiol.* **184**, 557 (1956)
- 25) KATZUNG, B., ROSIN, H. AND SCHEIDER.: *J. Pharmacol.* **120**, 324 (1957)
- 26) TRAUTWEIN, W. AND DUDEL, J.: *Pflüg. Arch. ges. Physiol.* **260**, 24 (1954)
- 27) HAJDU, S.: *Amer. J. Physiol.* **174**, 374 (1953)