EFFECTS OF CLONIDINE ON EXCITATION-CONTRACTION IN GUINEA-PIG TAENIA COLI

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Among the effects of clonidine [St 155, 2-(2,6-dichlorophenylamino)-2-imidazoline hydrochloride], those on the cardiovascular system have received considerable attention. Clonidine induces a biphasic change in blood pressure, consisting of initial brief hypertension and subsequent long-lasting hypotension (1). The initial hypertension has been explained by a sympathomimetic action of this drug and the subsequent hypotension by some centrally-mediated repression of the sympathetic tone (2-5).

The isolated preparation of taenia coli is an excellent material to study sympathomimetic and parasympathomimetic actions of drugs, responding to the former with inhibition and to the latter with excitation (6,7). The present paper reports the effects of clonidine on the excitation and contraction of taenia coli. The knowledge obtained with this material will be useful to interpret the drug actions on smooth muscle in general.

MATERIAL AND METHODS

All experiments were performed with taenia coli isolated from guinea-pig. The animal was sacrificed by a blow on the head. A piece of taenia coli, about 2 cm long in situ, was dissected free from the underlying tissue after the abdomen was opened and the colon exposed. The sucrose-gap method invented by Stämpfli (8) was used to record the electrical activity. The recording chamber employed in the present study was modified according to Washizu (9). Potential changes and isometric contractions were simultaneously recorded with a pen-writing oscillograph.

The Tyrode solution used was of the following composition (mM): NaCl, 137; KCl, 4; MgCl₂, 1.2; CaCl₂, 2.5; NaHCO₃, 12; NaH₂PO₄, 0.4; and glucose, 11. The solution with 20 or 80 mM K⁺ level was prepared by increasing KCl in an appropriate amount and by reducing equimolar NaCl without changing the concentrations of other salts. The Tyrode or test solution applied to the preparation was warmed up to 37 °C and aerated with a gas mixture of 95% O₂ and 5% CO₂. The drugs used were as follows: clonidine hydrochloride (C.H. Boehringer Sohn), atropine sulfate (Wako) and tetrodotoxin (Sankyo). These drugs were applied to the preparation by being freshly dissolved in the bath saline.

RESULTS

1. Effects of clonidine on spike discharge and contraction

Clonidine enhanced the rate and amplitude of spontaneous spike discharge of taenia...
FIG. 1. Effects of clonidine on electrical and mechanical activity of guinea-pig taenia coli. The first arrows (CL) indicate switchover to Tyrode solution containing clonidine (A, 0.3 mM; B, 1 mM) and the second arrows (W) readministration of normal Tyrode solution. In each set of records, the upper trace indicates contractile activity and the lower trace electrical activity registered by the sucrose-gap method.

coli. These effects were clearly demonstrated at a dose level of 0.3 mM (Fig. 1), although a level of 0.03 mM was only slightly effective. Administration of clonidine 1 mM resulted in oscillatory-type contractions and the discharge became burst-type. This tendency was observed with a lapse of time also after application of 0.3 mM. Recordings at a relatively high speed during such a contractile manner revealed increases in spike duration and contractile force, indicating better synchronous firing among fibers (Fig. 2). Further increase of clonidine level, i.e. to 3 mM, resulted in dissociation between the spike activity and the

FIG. 2. Electrical and mechanical activity of taenia coli before (A) and 18 min after (B) administration of clonidine 1 mM, recorded at a relatively high speed. Upper traces, contractile responses; lower traces, electrical potentials.
contractile machinery; namely, the development of tension associated with each spike became less.

It is known that clonidine makes some nonmyelinated nerve fire in a repetitive manner at relatively high dose levels (10). Therefore, a question may arise as to whether the effect of clonidine observed in the present study results from stimulating the cholinergic nerve, since the electrical and mechanical activity of taenia coli is elevated by acetylcholine (6). Effects of clonidine 3 mM were studied with a preparation treated with atropine 1 mM which was administered 1 min prior to the application of clonidine. As no inhibitory action of atropine was observed on the efficacy of clonidine, the changes were due to a direct action of clonidine on the muscle fibers without being mediated by nervous element (11). An additional piece of evidence for this view was obtained from experiments performed in the presence of tetrodotoxin 1.5 mM. This concentration of tetrodotoxin is considered to be sufficient to exclude nervous activity which might be involved, being about hundred times as great as the level which would be effective to suppress the nerve impulse (10). Clonidine (1 mM) showed an excitatory action in all of the six preparations treated with tetrodotoxin, the test responses being comparable with the controls taken without tetrodotoxin.

**FIG. 3.** Effects of clonidine on 20 mM potassium-induced responses of taenia coli. Top pair (A), control and test response with clonidine 0.3 mM; bottom pair (B), control and test response with clonidine 3 mM. In each set of records, the upper trace indicates contractile activity and the lower trace electrical activity. The first arrows indicate switchover to 20 mM K⁺ Tyrode solution without (K) or with clonidine (CL), and the second arrows (W) readministration of normal Tyrode solution.
2. Effects of clonidine on 20 mM potassium-induced response

Coupling between the spike discharge and contraction can be better studied at a 20 mM K⁺ level (Fig. 3), since the rate of spike discharge is elevated at this K⁺ level (12, 13). Clonidine 0.3 mM showed a tendency to increase the 20 mM potassium-induced contraction; the test response was 128.4 ± 14.5% (mean ± S.E., n=5, P=0.09) of the control taken without the drug. (The magnitude of the contraction in each series was defined by the average of the contractile response during a 10-min period starting 10 min after the K⁺ elevation. The same in subsequent series.) The results obtained with five different preparations showed a slight increase in spike amplitude in the simultaneously-recorded electrical activity of the test response, although a change in the rate of discharge was hardly detected. Elevation of clonidine concentration to 3 mM reduced the tension development in spite of the spike dis-

![Diagram](image)

**Fig. 4.** Effects of clonidine on 80 mM potassium-induced responses. Top pair (A), control and test response with clonidine 0.3 mM; bottom pair (B), control and test response with clonidine 3 mM. In each set of records, the upper trace indicates contractile activity and the lower trace electrical activity. The first arrows indicate switchover to 80 mM K⁺ Tyrode solution without (K) or with clonidine (CL), and the second arrows (W) readministration of normal Tyrode solution.
charge continuing without noticeable diminution in frequency and amplitude.

3. Effects of clonidine on 80 mM potassium-induced response

It is known that the mechanical response of taenia coli to such a high potassium level as 80 mM consists of initial phasic and ensuing tonic phases. The tonic contraction results from a steady depolarization, and the coupling between the depolarization and the contractile system is Ca²⁺ and metabolism-dependent (14, 15). Clonidine was scarcely effective in altering the 80 mM potassium-induced response at a 0.3 mM level. A dose level of 3 mM was capable of inhibiting the tonic phase in spite of the presence of the sustained depolarization which is comparable in amplitude with that of the control response (Fig. 4). Average values (mean ± S.E., n = 5) for the 80 mM potassium-induced depolarization and contraction were as follows: 22.7 ± 1.8 mV and 2.8 ± 0.4 g for the control and 21.3 ± 1.9 mV and 0 g for the test response obtained with clonidine 3 mM, respectively. The phasic response induced by 80 mM K⁺ was almost unchanged by clonidine 0.3 or 3 mM, showing a difference in resistivity between the phasic and tonic responses.

DISCUSSION

Clonidine enhanced the rate of spike discharge and increased the contractile force of the guinea-pig taenia coli, although relatively high dose levels were needed. The spike activity of guinea-pig taenia coli is very sensitive to α-sympathomimetic drugs, responding with inhibition of spike discharge and relaxation (7). The present study failed to demonstrate such an inhibitory action of clonidine at normal and 20 mM K⁺ levels. This observation is in contrast to the results obtained with other organs or tissues on which clonidine has some sympathomimetic actions (1-5, 16, 17). Furthermore, the clonidine effect was direct on the smooth muscle of taenia coli, differing from such nerve-mediated actions of the drug as reported for the negative chronotrophic action which can be blocked by atropine or vagotomy (16, 18).

It is generally considered that the potassium-induced contracture is metabolism-dependent and mediated by β receptors (19, 20). Whether or not β receptors are involved in the present effect of clonidine is still open for further study. However, the decoupling effect of clonidine observed at a 3 mM dose level is explained by the inhibition of the Ca²⁺ activity which mediates the excitation-contraction coupling (14, 21). The counteraction of Ca²⁺ on the inhibitory effect of clonidine on 80 mM potassium-induced contracture of taenia coli is already known (22).

SUMMARY

Effects of clonidine on the coupling between the electrical and mechanical activity of the isolated preparation of guinea-pig taenia coli were studied by the sucrose-gap method. Clonidine enhanced the spike discharge and augmented the contractile force at dose levels of 0.03-1 mM, showing no α-sympathomimetic action. Further analyses of clonidine effects on the coupling between the spike discharge and contraction and between the steady depolarization and contraction were made by means of 20 and 80 mM potassium-induced
responses. Clonidine showed a decoupling effect at a dose level of 3 mM.

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