THE INFLUENCE OF CERTAIN DRUGS ON THE RESPONSE OF SMOOTH MUSCLE TO DIRECT ELECTRICAL STIMULATION

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Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny,
Vol. 54, No. 12, pp. 57-60, December, 1962
Original article submitted January 16, 1962

Despite the extensive application of a number of drugs for the relief of spasm of the smooth muscle elements of the viscera, the mechanism of their spasmolytic action remains unknown.

In the present work we have studied the influence of a number of substances on the response of isolated smooth muscle to direct electrical stimulation.

The products we investigated included myotropic substances, 2,4-dinitrophenol, and substances which influenced specific biochemical systems. We also studied the influence of the change of the ionic composition of the solution bathing the muscle (absence of KCl) on direct electrical excitability.

EXPERIMENTAL METHODS

We used smooth-muscle segments of taenia coli from guinea pigs.

Direct electrical stimulation was applied by square-wave pulses of amplitude 3 V, frequency 20 cycles, and duration 10 mseconds. The stimulating electrode was insulated except for the tip, and was made of nichrome wire of diameter about 50 μ inserted into the muscle. The indifferent electrode was immersed in Tyrode's solution which bathed the muscle. We have previously described the mechanical recording devices [2].

When the Tyrode's solution contained no KCl, an equivalent amount of NaCl was added; control experiments showed that this exchange did not alter the mechanical properties of the smooth muscle.

All the substances to be investigated were added directly to the solution bathing the muscle.

EXPERIMENTAL RESULTS

Contraction of the smooth muscle induced by direct electrical stimulation lasted only during the stimulus period. In certain experiments, the smooth muscle reacted to direct electrical stimulation by relaxation; this effect usually occurred when muscle tone was raised.

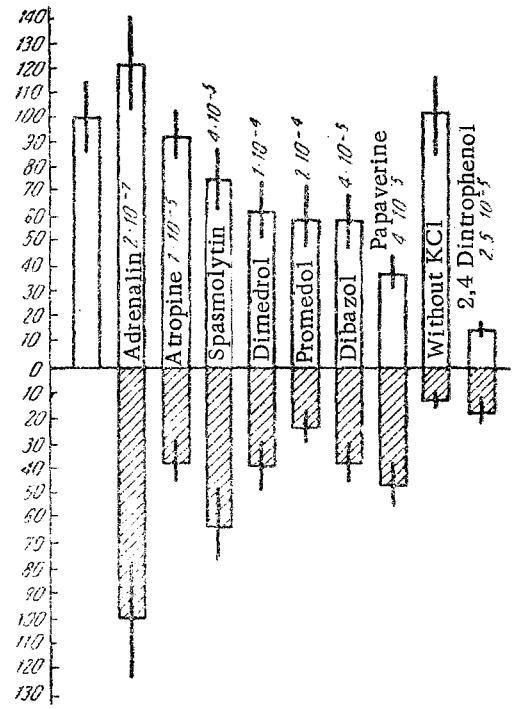
The extent of the contraction depended on the amplitude of the stimulus and on the duration of the individual pulse (the optimal pulse duration was 10 mseconds or more). A change of frequency from 20 to 100 cycles had no appreciable influence on the response.

The figure shows the influence of the substances investigated on the height of the contraction.

Despite the extremely relaxed condition of the muscle, $2 \cdot 10^{-7}$ adrenalin or $1 \cdot 10^{-5}$ atropine did not reduce the response to direct electrical stimulation (the reduced amplitude of the contractions under the influence of atropine was not statistically significant). In the experiments with adrenalin there was some tendency for the response to be increased.

Spasmolytin ($4 \cdot 10^{-5}$) and dimedrol ($1 \cdot 10^{-4}$) suppressed the smooth muscle contraction by 26 and 39% respectively. It is however important to note that the concentrations used were many times higher than the value $1 \cdot 10^{-6}$ which blocks the action of both acetylcholine and histamine.

Promedol ($2 \cdot 10^{-4}$), dibazol ($4 \cdot 10^{-5}$), and papaverine hydrochloride ($4 \cdot 10^{-5}$) reduced the response by 42, 43, and 63% respectively. Although the effects of promedol and dibazol were not significantly greater than that of dimedrol and spasmolytin, there could be no doubt of the increased effect from papaverine.



Influence of drugs on the tone and response of smooth muscle to direct electrical stimulation. Ordinate: Below – mean values (percentage) of the relaxation of smooth muscle under the influence of the substances investigated (the relaxation of the muscle caused by adrenalin is taken as 100%); Above – amplitude of the muscular contractions in response to direct electrical stimulation applied during the action of the substances (amplitude of contraction of the muscle in normal Tyrode's solution taken as 100%). The mean values are given together with the confidence limits.

As can be seen from the figure, the absence of KCl from Tyrode's solution, maintained for 15 min, did not alter the response.

A study of the action of 2,4-dinitrophenol showed that exposure of the muscle to Tyrode's solution containing $2.5 \cdot 10^{-5}$ of this substance reduced the response by 86%.

In our experiments, evidently, the direct local stimulation of smooth muscle caused some indirect muscular stimulation as well as stimulation of the intramural nervous system of the taenia coli of the guinea pig. However, from the experiments with atropine which blocks excitation of the cholinergic nerves of taenia coli, it is evident that the basis of the excitation of the smooth muscle was direct electrical stimulation of the muscle fibers. Elimination of the total effect of the nervous component by means of atropine did not appreciably alter the response.

Relaxation of the smooth muscle caused by adrenalin or atropine was as a rule associated with hyperpolarization and stabilization of the membrane of the smooth muscle fibers [5, 6]. However, as can be seen from the results, the hyperpolarization did not prevent the direct electrical stimulus from effectively depolarizing the muscle fiber membrane [7]. We may therefore suppose that hyperpolarization of the membrane is the release mechanism for the relaxation of the muscle induced by adrenalin and atropine.

At the present time little is known about whether the hyperpolarization of the smooth muscle fiber membrane is the result of the action of these substances on the whole membrane, or on certain portions specifically sensitive to certain substances.

Hyperpolarization and stabilization of the cell membranes of smooth muscle [10] due to changes of the ionic composition of Tyrode's solution (absence of KCl) are also without effect on the contractile properties of smooth muscle.

Previously [2] we showed that hyperpolarization of the cell membranes occurs also when smooth muscle is relaxed by papaverine. However, as the results show, relaxation induced by papaverine differs from relaxation caused by adrenalin or atropine. As can be seen from the figure, papaverine and other myotropic substances differ from atropine and adrenalin in strongly preventing the contraction of smooth muscle in response to direct electrical stimulation. We cannot therefore hold to the belief that the basis of the muscular relaxation from papaverine is the change of polarization of the smooth muscle membranes.

Papaverine has no cholinolytic properties [9], although, as can be seen from the experiments with atropine, such properties could not change the response of the muscle to direct electrical stimulation.

In our study of the influence of papaverine on the biochemistry of smooth muscle we showed that it suppresses the absorption of oxygen by the muscle [8], and enhances the activity of creatinkinase [1]. These results are of particular interest in relation to investigations [4] in which it was shown that oxygen lack in the solution surrounding the muscle leads to loss of smooth muscle tone when there is a 50-80% reduction of the creatin phosphate in the muscle. The results we have given lead to the conclusion that in the mechanism of action of papaverine, not only is there

an influence on smooth muscle membrane polarization, but also destruction of the biochemical processes responsible for the energy of contraction of the smooth muscle fibers plays an important part.

Unlike papaverine, 2,4-binitrophenol prevents the resynthesis of ATP by lowering smooth-muscle tone; the demand of the muscle for oxygen is initially enhanced, and subsequently suppressed [3]. Therefore, both papaverine and 2,4-dinitrophenol interfere with the metabolism of the high-energy compounds, and in this way alter the contractile properties of smooth muscle.

The tendency for adrenalin to increase the response of smooth muscle to direct electrical stimulation is evidently due to the fact that the smooth muscle of taenia coli increases in length while its contractile power remains unchanged. It is also possible that the increased formation in the muscle of lactic acid under the influence of adrenalin [11, 12] could lead to an increased response.

SUMMARY

Neither the absence of KCl from Tyrode's solution or the presence of $2 \cdot 10^{-7}$ adrenalin or $1 \cdot 10^{-5}$ atropine altered the response of the smooth muscle of a section of guinea pig taenia coli to direct electrical stimulation. The response was depressed by 26, 39, 42, 43, 63, and 86% through the action of $4 \cdot 10^{-5}$ spasmolytin, $1 \cdot 10^{-4}$ dimedrol, $2 \cdot 10^{-4}$ promedol, $4 \cdot 10^{-5}$ dibazol, $4 \cdot 10^{-5}$ papaverine, and by $2.5 \cdot 10^{-5}$ 2,4-dinitrophenol respectively. From published reports and our own results the explanation of the spasmolytic effects of adrenalin and atropine in smooth muscle appears to be that they act by an alteration of the condition of polarization of the smooth muscle fiber membranes, whereas papaverine appears to act both on the cell membrane and on the biochemical reaction of smooth muscle.

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