REACTIONS OF ARTERIOLES OF THE FROG SUBMAXILLARY MUSCLE TO STATIC AND PERIODIC DISPLACEMENTS AND CONTRACTIONS OF ITS FIBERS

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Low-frequency (about 4/sec) oscillatory displacements of resting fibers of the frog submaxillary muscle, leading to bending microdeformations of the arterioles located among them, cause considerable dilatation of them [3]. This phenomenon is evidence of the essential role of mechanical factors in working hyperemia of skeletal muscles [4]. During static displacement of a group of muscle fibers, the arterioles located among them must also undergo microdeformation. However, it is not known whether such displacement does in fact affect arterioles.

The aim of the experiments described below was, first, to establish whether aerterioles of a skeletal muscle respond to static displacement of its fibers and, second, to compare the effect of such a procedure with responses of the same arterioles to oscillatory displacements of muscle fibers and also to their contraction.

EXPERIMENTAL METHOD

The submaxillary muscles of 9 frogs, anesthetized with viadril (3.7 mg intravenously), were exposed as described previously [2, 3]. The submaxillary branches of the V and VII cranial nerves were divided and the peripheral end of the branches of both trigeminal nerves were placed on unipolar electrodes. The parameters of the stimuli (duration 10 μsec, frequency 4 Hz/sec, amplitude 3 times the threshold strength for excitation of motor fibers) were chosen beforehand [5]. Arterioles with clearly distinguishable walls were selected under the microscope and examined in transmitted intermittent light. This type of illumination prevents suppression of myogenic tone of the arterioles and maintains their reactivity at a high level [2]. To displace the muscle fibers a piezoelectric crystal plate, to which a microsuction device consisting of a glass tube, the tip of which was drawn out into a cone 0.1-0.25 mm in diameter, was securely fixed, was used. By means of a micromanipulator the tip of the suction device was brought up to an area of muscle containing only capillaries, to a distance of 0.15-1 mm of the test arteriole, and a vacuum was created in it sufficient to ensure reliable attachment to the muscle fibers [3]. If a constant tension was applied to the piezoelectric crystal plate it bent parallel to the muscle surface, and the force was transmitted through the suction device to a group of muscle fibers. Under these circumstances they were displaced in the direction of bending of the plate, causing microdeformations of the microvessels located among them. The order of conduct of the experiment was as follows. Having fixed the suction device and waited 30-40 min, photomicrographs of the test arterioles were obtained every 30 sec, usually for 8-10 min. The camera was switched to the continuous film delivery mode [5] 15 sec before the beginning of stimulation, and the frequency of photomicrography was increased to 4/sec. The state of the vessel was recorded at the same frequency on moving photographic film for the next 90 sec, during which static displacement of the muscle fibers was maintained, and then for a further 15 sec after the end of the procedure. The photomicrographs were subsequently obtained less frequently: every 30 sec or 5-9 min. Responses of the same vessel to oscillatory displacement of the muscle fibers (frequency 4/sec) and also to low-frequency (about 4/sec) interrupted tetanus, were recorded by the same scheme. In both cases stroboscopic illumination was used: the light pulses were strictly coordinated with application of electrical impulses to the piezoelectric crystal plate or to the nerves of the muscle [5]. Values of the constant or saw-

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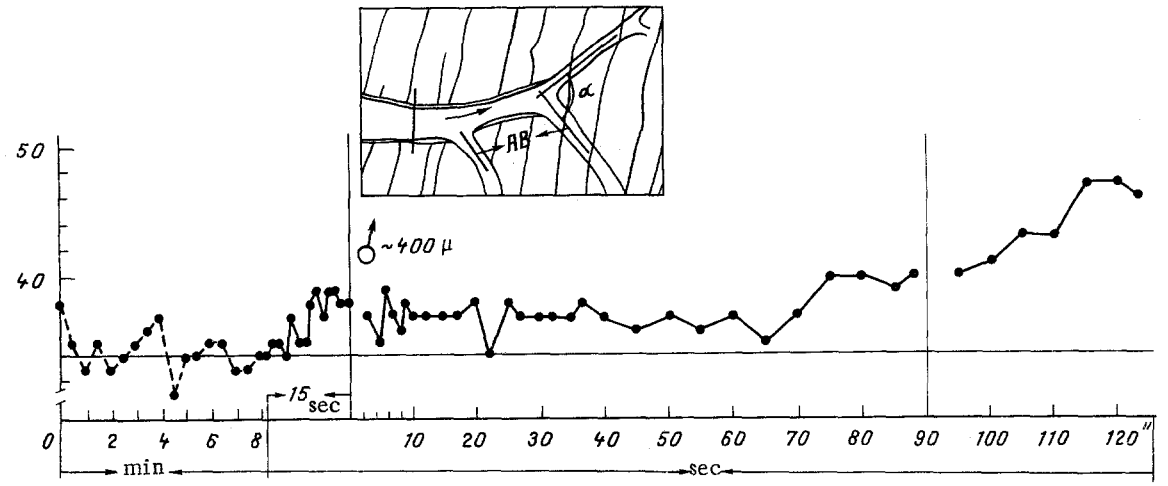

Fig. 1. Changes in diameter of arteriole before, during, and after static displacement of fibers of submaxillary muscle surrounding it. Position of arteriole relative to muscle fibers shown diagrammatically, arrow inside vessel indicates direction of blood flow, transverse line shows place where diameter was measured. Circle with arrow outside diagram shows direction of displacement of fibers, and number gives distance from suction device to arteriole. During static mechanical displacement arteriole was shifted by 150 μ relative to its initial position. Under these circumstances, the distance between branches (AB) was increased by 10 μ and the angle was reduced by 14°. Abscissa, time; ordinate, diameter (in μ). Vertical lines denote period of mechanical action, horizontal line corresponds to diameter of vessel averaged for period before procedure.

tooth voltage applied to the piezoelectric plate to create static or oscillatory displacements of the muscle fibers respectively were chosen so that the arteriole was moved through approximately the same distance, similar to its displacement during muscle contraction. The external diameter of the arterioles was measured from photomicrographs. The results of only those experiments in which the pressure measured in the dorsal aorta was not less than 30 cm water were taken into account.

EXPERIMENTAL RESULTS

Altogether 12 arterioles were subjected to static displacement 17 times. The arterioles were moved through 0.15-0.27 mm relative to their initial position. These values are comparable with the constant component of displacement of the submaxillary muscle in response to stimulation of the homonymous symmetrical nerve with a frequency of 4-6 Hz/sec [11]. During displacement of the arterioles the angles of divergence of the lateral branches and terminal bifurcations and also the distance between the ramifications of the vessels change (Fig. 1). Arterioles of resting muscle exhibit vasomotion — spontaneous changes of diameter (Figs. 1-3). To discover the response of an arteriole to the action of a certain factor, the values of the changes induced in its diameter must be related to its initial diameter, average for the period of observation before the procedure. The initial mean values of the diameter of different arterioles ranged from 20-55 μ. In the dilator phase of vasomotion the diameter increased by 5-43% compared with its mean value, and in the constrictor phase it decreased by 5-36%.

The mean values of the diameter of the arteriole, the result of displacement of which is shown in Fig. 1, were 34.4 ± 3.6 μ ($m \pm \sigma$) initially, and 37.2 ± 3.4 μ during static displacement, i.e., this did not lead to a statistically significant increase in diameter ($p = 0.05$). The result for eight arterioles (13 tests) was the same: values of their diameter averaged for the whole period of static displacement did not differ statistically significantly from the original mean values ($p > 0.05$). Only for three arterioles (3 tests) was the mean increase in diameter during static displacement 12, 19, and 26% relative to its original mean value ($p \leq 0.02$). Admittedly, detailed analysis showed that although one of these arterioles, after only 6 sec of displacement, was wider than at the maxima of the

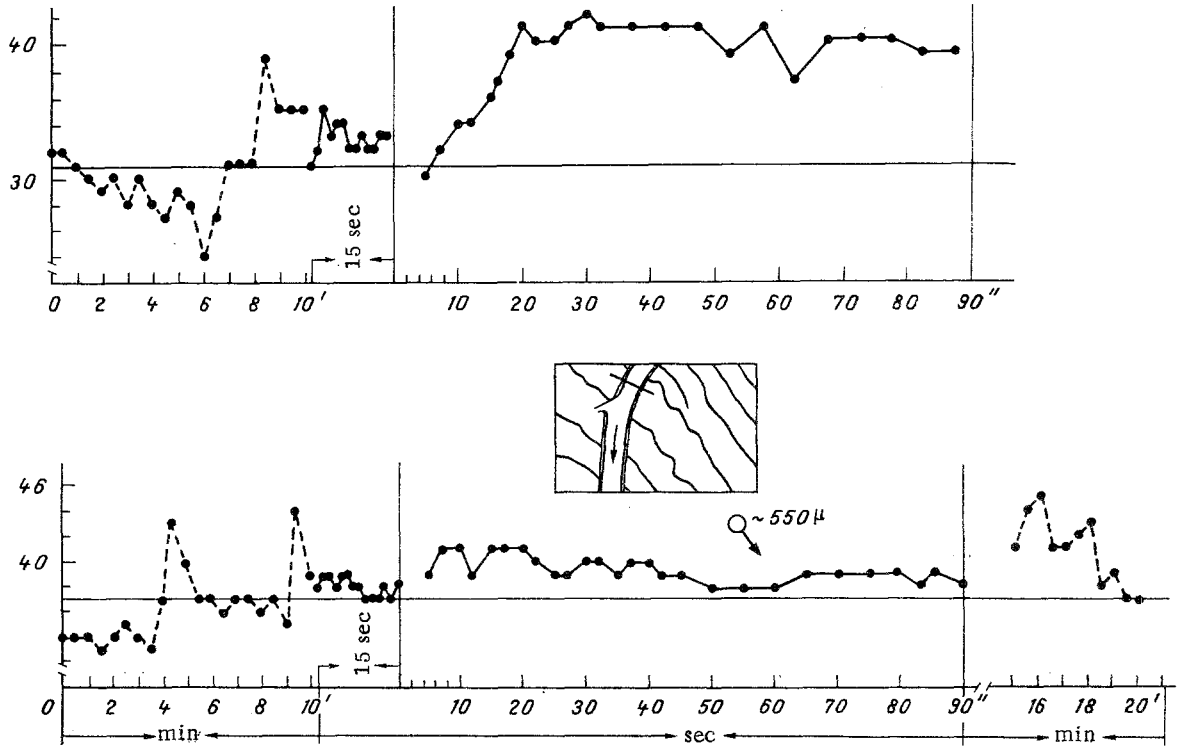

Fig. 2. Different behavior of the same arteriole during two equal static displacements of muscle fibers. Displacement of arteriole relative to its initial position 250 μ. Legend as to Fig. 1.

dilator phases of vasomotion, starting with the 3rd second its dilatation steadily began to become narrower. Dilatation of the other two arterioles was inconstant: in one case it was observed only during the first of two equal displacements (Fig. 2), in the other only during the first of two equal displacements (Fig. 2), in the other only during repeated displacements. Finally, one arteriole began to constrict even before the beginning of static displacement of its fibers and this process was intensified during displacement, so that toward its end the diameter was 22% less than its average initial value. During repeated and somewhat greater displacement, which also began during the constrictor phase of vasomotion, the diameter of this arteriole remained within the limits of its initial value throughout the period of displacement.

Static displacement of muscle fibers thus usually does not lead to dilatation of the arterioles. In those few cases when they appeared to be wider than at the maximum of the dilator phases of the initial vasomotion, their dilatation was slight and transient. Such short and weak dilatation of the arterioles is most probably only a somewhat strengthened dilatation of the arterioles is most probably only a somewhat strengthened dilator phase of vasomotion, and in one case, a deepened constrictor phase. The negative result — actual absence of response of the arteriole to static displacement of their surrounding muscle fibers — cannot be ascribed in any way to low reactivity of the vessels. During oscillatory displacement of the fibers these same arterioles, shifted relative to their initial position by virtually the same amount, were invariably dilated. On average the increase in diameter of 11 arterioles in this case reached 36% compared with the mean initial value. The increase in diameter for 11 arterioles during contraction of the muscle varied from 32 to 69% (Fig. 3).

The grounds for undertaking the experiments described here and previously [3] were the histomechanical hypothesis of working hyperemia of skeletal muscles [4], according to which arterioles are "bound" by fibers of the adventitia to the perimysium of the muscle bundles, as a result of which these vessels are tightened along their length, and this tension maintains the myogenic tone of the arterioles. It has been suggested [4] that during displacement of muscle fibers in the same direction as during contraction, longitudinal tension of the arterioles and, at the same time, their myogenic tone, are reduced and the vessels dilated. However, experiments showed that the arterioles are dilated in response only to oscillatory displacements of the muscle fibers (Fig. 3). The myocyte membrane of the arteri-

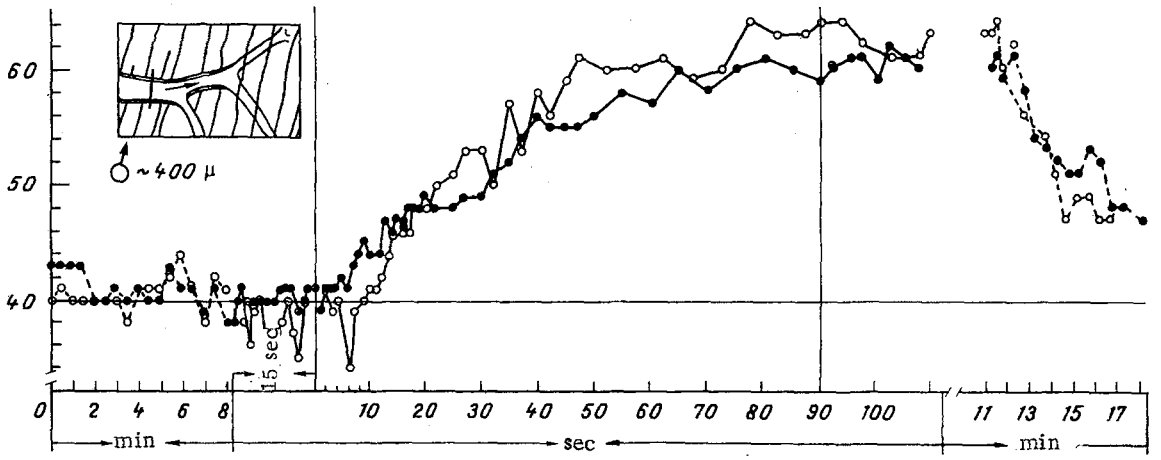

Fig. 3. Dilatation of arteriole (the same vessel as in Fig. 1) during periodic (4/sec) displacement of muscle fibers (filled circles) and during their contraction at the same frequency (empty circles). Displacement of arteriole during periodic mechanical action 150 μ; during contraction of muscle 180 μ. Legend as to Fig. 1.

oles thus responds only to the velocity (dynamic) component of the reduction of its stretching, and an essential condition for relaxation of myocytes is the ability of their membranes and (or) of certain intracellular processes, triggering on weakening of tension of the arteriole, to produce summation of the effect thus arising.

So far as this predominant sensitivity of vessels to the dynamic component of stretching is concerned, this is a property possessed by the portal vein [8, 11]. During passive shortening (due to elastic properties) of the previously stretched vein the discharge frequency of its pacemakers falls more strongly (sometimes to zero) than after recovery of the initial length of the vein. The lowering of the discharge frequency of the pacemaker cells during passive shortening of the vessel can be explained by weakening of the preliminary stretching of these cells [8], but it may perhaps be due to the "weak" deformations of their membrane, leading to its hyperpolarization. Indirect evidence in support of this view is given by relaxation of a region of arterioles in response to its "weak" compression by relaxation of a region of arterioles in response to its "weak" compression and local contraction in response to stronger compression [7]. These phenomena can be compared with responses of the squid giant axon membrane: it is hyperpolarized by comparatively weak tangential strain and depolarized in response to stronger strain [12].

During oscillatory displacements of muscle fibers the phases of their displacement and return to the initial position lasted about 50 and 200 msec [3]. It may appear surprising that such short procedures should be capable of altering the state of the arterioles. We know that a long after-effect, lasting several seconds, is characteristic of processes arising even during short (of the order of milliseconds) mechanical actions on the nerve fiber membrane [6, 9, 13]. The effectiveness of oscillatory displacements of arterioles (Fig. 3) suggests that ability to maintain effects induced by short mechanical actions for a long time, and ability to carry out summation of these effects are also properties of the arteriolar myocyte membrane.

Both depolarization and hyperpolarization of the membranes during stretching have been explained until recently by an increase in ionic permeability [6, 9, 13]. It is more likely however, that mechanical stimulation is perceived by special ionic channels activated by stretching of the membrane [10], and that selective sensitivity to the rate of growth or reduction of deformation (in our case, to weakening of longitudinal tension of the arterioles) is determined by extra- and intracellular structures which act as mechanical filters, tuned to receive only the dynamic component of microdeformations of the arterioles.

LITERATURE CITED

1. L. A. Mirzadaeva and E. V. Lukoshkova, Byull. Éksp. Biol. Med., No. 4, 10 (1971).
2. R. S. Sonina, Fiziol. Zh. SSSR, 67, No. 9, 1348 (1981).
3. R. S. Sonina and V. M. Khayutin, Byull. Éksp. Biol. Med., No. 12, 657 (1986).
4. V. M. Khayutin, Physiology of Man and Animals [in Russian], Vol. 23, Moscow (1979), pp. 46-106.

5. V. M. Khayutin, R. S. Sonina, and I. K. Evstifeev, Fiziol. Zh. SSSR, 70, No. 5, 681 (1984).
6. G. Ganot, B. S. Wong, L. Binstock, and G. Ehrenstein, Biochim. Biophys. Acta, 649, No. 2, 487 (1981).
7. R. T. Grant, Heart, 15, 257 (1930).
8. B. Johansson and S. Mellander, Circulat. Res., 36, No. 1, 76 (1975).
9. F. J. Julian and D. E. Goldman, J. Gen. Physiol., 46, No. 2, 297 (1962).
10. F. Sachs, Membr. Biochem., 6, No. 2, 173 (1986).
11. S. Sigurdsson, J. Johansson, and S. Mellander, Acta Physiol. Scand., 99, No. 2, 183 (1977).
12. S. Terakawa and A. Watanabe, Pflügers Arch., 395, No. 1, 59 (1982).
13. J. B. Wells and D. E. Goldman, Biophys. J., 32, No. 2, 91a (1981).