ARTERIOLAR RESPONSES TO RHYTHMIC DISPLACEMENT OF FROG SUBMAXILLARY MUSCLE FIBERS

R. S. Sonina and V. M. Khayutin

UDC 612.133-06:612.741

KEY WORDS: arterioles; working hyperemia; microdeformation of arterioles; muscle fibers; frog.

Dilatation of arterioles of skeletal muscles during work was demonstrated more than 100 years ago [1, 7], although this phenomenon was confirmed by direct microscopic studies only quite recently [3, 4, 6, 8, 11]. However, it is not yet known by what concrete mechanism contraction of muscle fibers causes relaxation of the myocytes of arterioles [5]. According to the histomechanical hypothesis of functional hyperemia, this relaxation may be the result of mechanical effects of contracting fibers on the arterioles [5]. It is suggested that as a result of changes in configuration of the arterioles the tone of their smooth muscles may be reduced. In turn, this may reduce the frequency of spike generation by the pacemakers [15], and a change in the mutual arrangement of the smooth muscles could impede the myogenic conduction of potentials from the pacemakers to the driven cells [10]. All these events could lead to relaxation of the arteriolar myocytes and thus increase the diameter of the vessels. Just as in [14], we apply the term arterioles to intramuscular arterial vessels whose tunica media consists of one or two layers of smooth-muscle cells.

The aim of this investigation was to determine whether rhythmic local displacement of fibers of a resting skeletal muscle can produce changes in configuration of the arterioles and lead to their dilatation.

EXPERIMENTAL METHOD

The submaxillary muscles were exposed and decentralized in nine frogs, anesthetized with Viadril (3.7 mg, intravenously), by dividing the submaxillary branches of the V and VII cranial nerves [6]. A region of an arteriole located on the surface of the muscle was chosen for investigation under the MBI-6 microscope in transmitted light from an ISSh-100-3M flash tube [6]. To change the configuration of this region, and thus to cause microdeformation of its walls, a group of muscle fibers, among which this arteriole was located, was displaced in the direction in which they shorten during natural contraction. The force required to displace the fibers was created by means of a piezocrystal plate (60 × 15 × 0.7 mm; bending of the free end by 2 μ per volt). To transmit the bending movement of the plate, arising in response to application of a voltage to it, one end of it was firmly secured to a manipulator (MO-103, Narishigi), and a glass tube 1.2 mm in diameter was fairly fixed to its free end. As a preliminary step, the end of the tube facing the muscle was drawn out in a flame into a cone, the diameter of which varied from 100 to 250 μ. This cone served as a suction device. The end of the tube fixed to the plate was connected by a flexible tube to a syringe, filled with mineral oil. Under low power of the microscope (24.5 ×) the suction device was applied to the surface of the muscle at distances of 0.3 to 0.9 mm from the chosen region of the vessel and placed in firm contact with the surface. By moving the plunger of the syringe by means of a microscrew, a vacuum was created in the glass tube; under these circumstances the perimysium was drawn into the cavity of the suction device, which was thus fixed to the group of muscle fibers. Bending of the piezocrystal plate caused displacement of the muscle fibers. The amplitude of bending of the plate, and consequently, the amplitude of displacement of the fibers and the degree of deformation of the arterioles could be altered by varying the voltage applied to the piezocrystal plate up to 220 V. The source of voltage was a generator (developed by I. K. Evstifeev), which enabled the plate to be bent either continuously or periodically. In

Laboratory of Biomechanics and Regulation of the Circulation, All-Union Cardiologic Scientific Center, Academy of Medical Sciences of the USSR, Moscow. (Presented by Academician of the Academy of Medical Sciences of the USSR V. N. Smirnov.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 102, No. 12, pp. 657-661, December, 1986. Original article submitted April 29, 1986.

0007-4888/86/0012-1637$12.50

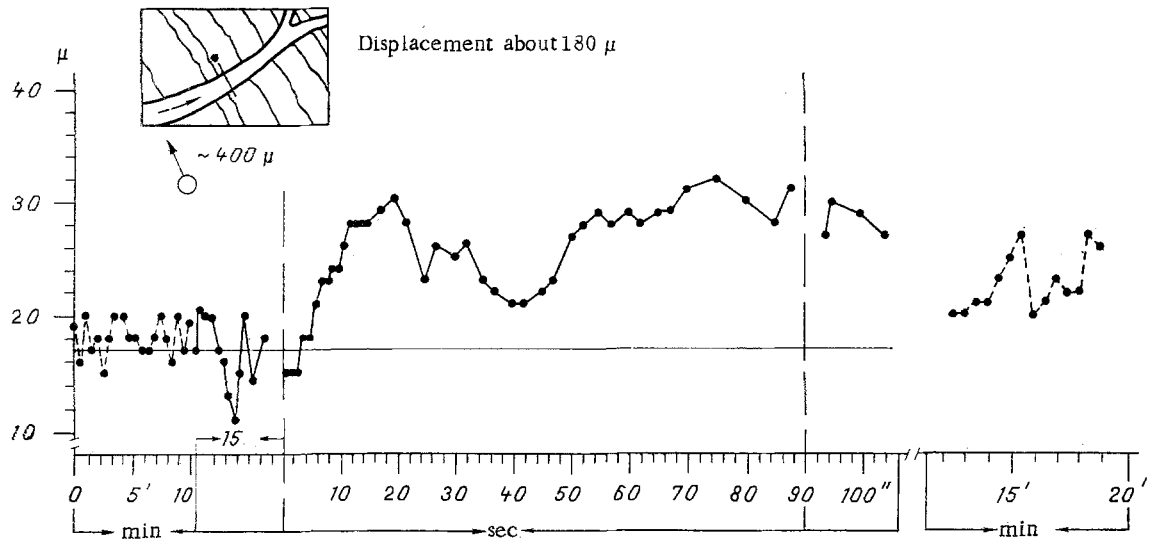

Fig. 1. Changes in diameter of arteriole of submaxillary muscle before, during, and after rhythmic local displacement of its fibers. Abscissa, time; ordinate, diameter (in μ). Vertical broken lines denote period of mechanical displacement, horizontal line corresponds to diameter of vessel averaged for the resting period. Inset: diagram of photomicrograph of arteriole (direction of blood flow indicated by arrow, place where diameter was measured shown by line across it). Arrow outside frame shows direction of displacement of fibers, and numbers indicate distance from suction device (empty circle) to arteriole and magnitude of displacement.

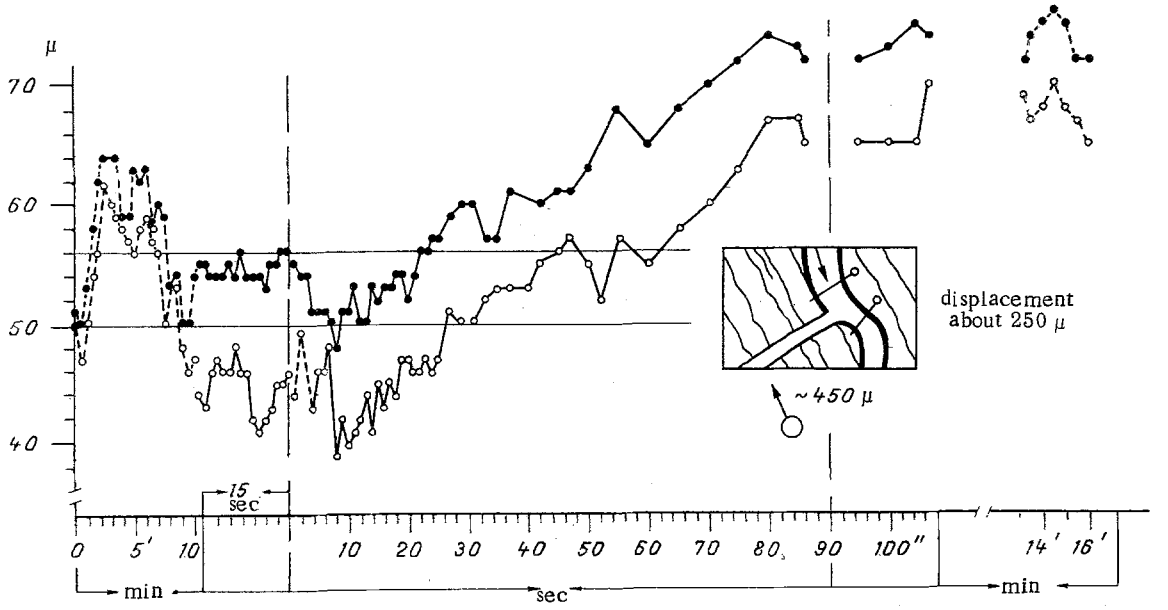

Fig. 2. Dilatation of arteriole during displacement of muscle fibers (beginning of dilatation delayed, probably by development of constrictor phase of vasomotor activity). Graph shows changes in diameter of two regions of an arteriole located about 100 μ apart (scheme). Remainder of legend as to Fig. 1.

the latter case, the voltage on the plate reached a maximum in the course of 50 msec, and then fell to zero in 200 msec. These durations of the fronts of the electric pulses correspond to some degree to the duration of phases of contraction and relaxation of the submaxillary muscle during stimulation of its motor fibers with a frequency of 4 Hz. To estimate the periods and range of spontaneous vasomotor contractions of the arteriole, the region chosen was photographed every 30 sec for 7-9 min. Next, 15 sec before the beginning of displacement of the muscle fibers the state of the vessel was recorded on motion picture film with a frequency of 4 frames/sec [6]. Next, pulses were applied with the same frequency for 90 sec to

the piezocrystal plate. Every triggering pulse of the voltage generator thus triggered the power unit of the ISSh-100-3M flash tube, so that the state of the arteriole could be recorded in identical phases of rhythmic displacement of the muscle fibers, namely at the times of restoration of their original length. After displacement of the fibers had ceased, the vessel continued to be photographed with a frequency of 4 frames/sec for a further 15 sec, and thereafter less frequently — every 30 sec for 5-6 min. The state of the animals' cardiovascular system was judged by the blood pressure, measured in the dorsal aorta by means of an electromanometer, and recorded on a KSP-4 potentiometer. It varied from 30 to 40 cm water. The diameter of the arterioles was measured in serial photomicrographs (total magnification 101×).

EXPERIMENTAL RESULTS

Displacement of the chosen region of the vessel was measured with an ocular micrometer under low power (24.5 ×). This displacement, during changes in length of the muscle fibers with a frequency of about 4 Hz, varied from 0.05 to 0.25 mm for different arterioles. During displacement of the muscle fibers the configuration of the arterioles located among them changed. They were bent mainly in the direction of the optical axis of the microscope. However, since the photographs were taken at the times of restoration of the initial position of the muscle fibers, a very small correction for sharpness of the image was needed.

All 16 regions of 11 arterioles chosen for study responded to rhythmic mechanical stimulation by dilatation (Figs. 1-3). At the peak of the dilator response the diameter of the different arterioles increased from 16 to 126% relative to the mean value over a period of time (the mean was calculated over a period of 7-9 min before mechanical stimulation). The amplitude of the dilator response to displacement of the muscle fibers always exceeded the amplitude of the spontaneous increases in diameter of that same microvessel, i.e., of the dilator phases of vasomotor activity (Figs. 1-3). The external diameter of the different arterioles (averaged over a period of 7-9 min) varied from 17 to 56 μ; at the peak of the dilator phases of vasomotor activity, moreover, it could be increased by 5-89%, and at the peak of the constrictor phases, it could be reduced by 3-47%.

At the beginning of mechanical stimulation the arterioles were in different phases of vasomotor activity. The marked difference in latent period of their dilator response to rhythmic changes of configuration may be linked with this fact. The latent period for five arterioles was 3-8 sec (Fig. 1), for the rest between 10 and 32 sec (Figs. 2 and 3). In four cases dilatation of the microvessel was preceded by a decrease in its diameter, either rapid or delayed a little compared with the beginning of displacement of the muscle fibers (Fig. 2). Dilatation of most arterioles reached a maximum in the course of 50-80 sec, but for the five vessels which began to dilate after a relatively short latent period, the maximum of the response was reached more quickly — after 13-40 sec (Fig. 1).

In one experiment responses of the same region of an arteriole were recorded to three consecutive identical mechanical stimulations (Fig. 3). In every case the latent period of the dilator reactions, the time taken to reach the maximum, and the magnitudes of the responses were virtually identical.

It was shown by Mirzadaeva [2], and later by others, that during tetanic contraction of a skeletal muscle the configuration of its arterial microvessels changes, and most frequently they bend. Meanwhile arterioles have high mechanical sensitivity, for they dilate in response to light tough, and constrict in response to strong pressure [9]. These observations suggested that changes in configuration of arterioles, causing microdeformation of their walls, may lead to weakening of tone of the myocytes [5]. However, the results of the only attempt so far made to induce dilatation of arterioles, by stretching a small bundle of muscle fibers of the hamster cremaster muscle with a frequency of about 4 Hz, were negative [8]. The positive results of our own experiments is evidently due to two circumstances: 1) the higher reactivity of the microvessels, as shown by preservation of their vasomotor activity; 2) the greater region of mechanical stimulation of the arterioles and, probably, the greater displacement of the muscle fibers.

During rhythmic displacement of muscle fibers the vasomotor activity of the arterioles may perhaps not be completely blocked. In that case, transient constriction of a microvessel, already dilated to the limit, which sometimes took place despite continuing mechanical stimulation, may be linked with preservation of vasomotor activity (Fig. 1). It is difficult to say what causes the initial constriction of some vessels (Fig. 2): whether it is deepening

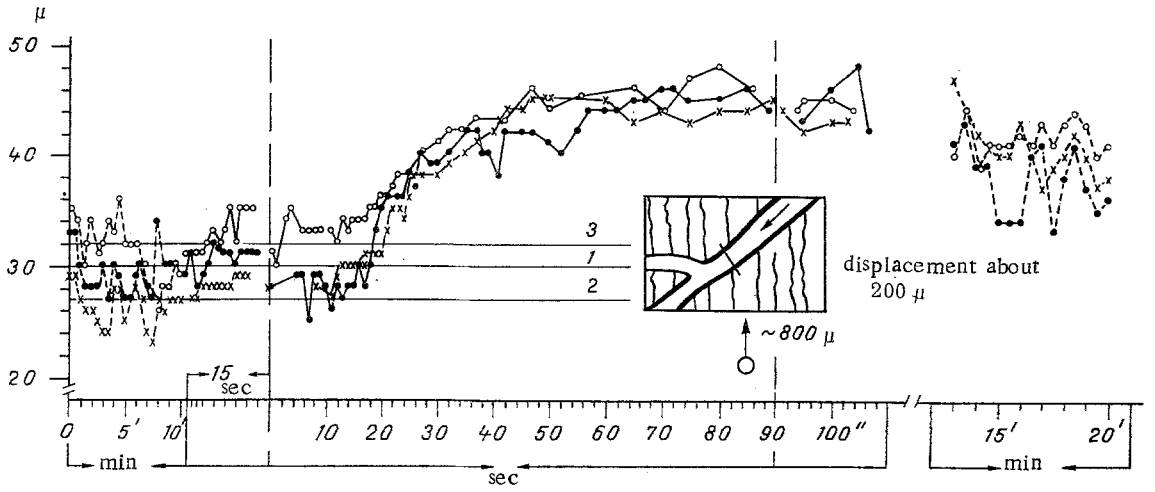

Fig. 3. Dilatation of arteriole during three identical repeated mechanical stimulations. Filled circles — first stimulation; crosses — second, empty circles — third stimulation. Horizontal lines show values of diameter averaged for resting periods before first, second, and third stimulations (numbers above lines). Remainder of legend the same as in Fig. 1.

of the constrictor phase of vasomotor activity initiated previously or the result of some local concentration of forces which, in the case of stronger mechanical action on the arterioles [9], could cause contraction of their muscular coat.

When the present experiments were planned it was assumed that mechanical stimulation has a direct action on arteriolar myocytes [5]. Meanwhile mechanical stimulation can accelerate certain biochemical processes in skeletal muscles. If muscles incubated in vitro are stretched periodically, this almost doubles the rate of synthesis of some proteins, increases the ATP concentration, and accelerates glucose uptake [12]. This acceleration of metabolism is considered [13] to be linked with the effect of mechanical stretching on prostaglandin release and on the intracellular Ca^{++} distribution. However, the processes accelerated are mainly anabolic, and, what is particularly important, they proceed slowly. The existence of these mechanically sensitive biochemical reactions cannot therefore compel rejection of the idea that arterioles dilate as a result of direct mechanical stimulation of their myocytes. Whatever the case, the essential fact is that dilatation of arterioles of this order in response to rhythmic displacement of resting muscle fibers reaches the same magnitude as during serrated tetanus with a frequency of about 4 Hz [4, 6]. Values of other parameters of accompanying reactions were closely similar: the latent period of dilatation of the vessels and the time taken to reach its maximum. Often the increase in diameter of arterioles was preceded by a decrease in diameter, both during contraction of the muscles [4, 6] and during mechanical displacement of its resting fibers (Fig. 2). This suggests, in the writers' opinion, that the mechanical factor, namely microdeformation of the arterioles, plays an essential role in the working hyperemia of skeletal muscles.

LITERATURE CITED

1. V. K. Zadler, "On the circulation in resting, contracted, and fatigued muscle of a living animal," Dissertation, Khar'kov (1875).
2. L. A. Mirzadaeva, "Development of methods and some results of intravital study of the microcirculation in contracting skeletal muscle," Candidate's Dissertation for the Degree of Medical Sciences, Moscow (1971).
3. R. S. Sonina, Regulation of the Circulation in Skeletal Muscles [in Russian], Riga (1980), pp. 127-137.
4. R. S. Sonina and O. Ya. Kaufman, Byull. Éksp. Biol. Med., No. 7, 107 (1984).
5. V. M. Khayutin, Progress in Science and Technology. Series: Human and Animal Physiology [in Russian], Vol. 23, Moscow (1979), pp. 46-106.
6. V. M. Khayutin, R. S. Sonina, and I. K. Evstifeev, Technical Aspects of the Use of Television Microscopy to Investigate the Cardiovascular System [in Russian], Leningrad (1982), pp. 32-36.

7. W. H. Gaskell, J. Physiol. (London), 3, 48 (1880).
8. R. J. Gozczynski, B. Klitzman, and B. R. Duling, Am. J. Physiol., 235, 494 (1978).
9. R. T. Grant, Heart, 15, 257 (1930).
10. G. D. S. Hirst and J. O. Neild, J. Physiol. (London), 280, 87 (1978).
11. J. M. Marshall and H. C. Tandon, J. Physiol. (London), 350, 447 (1984).
12. R. M. Palmer, P. J. Reeds, G. E. Lobley, and R. H. Smith, Biochem. J., 198, 491 (1981).
13. R. M. Palmer, P. J. Reeds, and R. H. Smith, J. Physiol. (London), 332, 13P (1982).
14. J. A. G. Rhodin, Handbook of Physiology, Section 2, The Cardiovascular System, Vol. 2, Washington (1980), pp. 1-31.
15. E. Zelcer and N. Sperilakis, Blood Vessels, 19, 301 (1982).