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Further observations on the blood vessels of skeletal muscle (rat cremaster)

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INTRODUCTION

Reference is made in a previous paper (Grant, 1964) to vascular channels that remain open to carry on the reduced circulation in the resting cremaster muscle. It was then uncertain whether or not the open vessels were of the same nature as the thoroughfare channels with a similar function described by Zweifach & Metz (1955) in the rat spino-trapezius muscle. The latter vessels are arteriolar and venular and lie in the cleavage planes of the muscle while the interior of the muscle is ischaemic. They form the central channels of recognizable functional vascular units and have a special structure. The arteriolar portion, the metarteriole, 10–20 μm diameter when dilated, is characterized by discontinuously arranged smooth muscle cells; beyond the metarteriole, the muscle cells progressively disappear and the channel becomes a venule (Chambers & Zweifach, 1944, 1947). In the cremaster (Grant, 1964), however, the channels through which the circulation persists in the resting state are within the muscle layers and lie parallel to the muscle fibres; their diameters are little if at all wider than those of neighbouring capillaries when these are dilated. Their structure could not be determined in the living preparation. Therefore, further observations have been made on the cremaster to determine more precisely what vessels remain open when the muscle is at rest, and whether these vessels form part of a vascular unit; and if so, whether they are structurally distinguishable. This has involved an examination of vessels during life and of the same vessels later displayed histologically by intravascular fixation and staining.

METHODS

The *general procedure* has been to study the vessels during life and to photograph them in both the resting and dilated state. The animal is then killed and the vessels fixed and stained in the dilated state by a perfusion technique.

The *method for observing the living vessels* has already been published (Grant, 1964, 1966). A minor modification is the omission of gelatin from the irrigating fluid. Improved illumination is provided by an 'intense lamp' (Vickers Instruments Ltd) with green and heat absorbing filters. A stereoscopic microscope with magnifications of 25 and 50 diameters is suitable.

For photography, a 35 mm camera with viewing telescope (Leica camera and Mikas micro attachment; E. Leitz Instruments Limited, London) is fixed to one eyepiece of the microscope. The body of the microscope is tilted so that the lens system

is vertical to the area of the cremaster viewed. Because of the curvature of the muscle and the different depths at which the vessels lie, it is seldom possible to focus sharply on all the vessels simultaneously. The slight respiratory movement necessitates brief exposure, attained by an electronic flash (Braun F26) also provided with a green filter.

The *method for preparing the cremaster for histological examination* previously described (Grant, 1966) requires modification for intramuscular staining. The method of intravascular staining now used is itself a modification of that devised for earlier work (Grant, 1929–31).

At the end of observation during life, 500 units heparin is injected into a catheter tied into the jugular vein. This is followed about 5 min later by 20 ml heparinized Ringer's solution (10 units/ml) injected slowly. During this injection a carotid artery is opened. This not only kills the rat but also allows much blood to escape, rendering the subsequent catheterization of the aorta cleaner. The administration of heparin is important because it prevents the formation of small clots which, without it, are liable to block small vessels. The thighs and base of the tail are ligatured tightly; this reduces the volume of tissue to be perfused and avoids the diversion of perfused fluid from capillaries through the numerous arteriovenous anastomoses in the feet and tail.

The apparatus for perfusion consists of a pressure reservoir connected on the one side to a sphygmomanometer bulb and a mercury manometer and on the other to a 500 ml bottle containing the fluid to be perfused. Since it is essential to avoid the entry of air into the vessels and yet allow the perfusing fluid to be changed easily, the bottle is connected to an air trap. This consists of a glass cylinder about 5 cm long by 2·5 cm wide, closed top and bottom with rubber bungs. The top is pierced by a tap and the inlet tube, the bottom by a tap and the outlet tube. The taps allow adjustment of the fluid level within the trap. The outlet tube, about 3 mm internal diameter, joins on to a short length, 5–10 cm, of polythene tubing, 0·58 mm internal diameter (PE 50, Clay Adams Inc., New York) for insertion into the aorta.

Procedure for perfusion. The abdomen is opened by a midline incision and the intestines tied off and excised. The aorta below the renal vessels is cleared. The abdominal cavity is washed out and filled with Ringer's solution and through this the polythene tube is tied into the aorta. Perfusion with Ringer's fluid is begun and the inferior vena cava opened. The cremaster (kept moist) is observed under the microscope to see that all blood is washed from the vessels. Sometimes red cells remain in a few vessels; they are cleared by gentle stroking with a glass rod. When the caval effluent is clear, 4 % neutral formaldehyde with 0·1 M sucrose is perfused. The Ringer's solution and the formaldehyde, filtered before use, are maintained at a temperature of 35–40 °C and under a manometer pressure of 160–200 mgHg. The corresponding lateral pressure in the polythene tube entering the aorta is 100–120 mmHg. Formaldehyde perfusion is continued for at least 2 h, during which time gross oedema develops. Perfusion is then interrupted until the following morning, the cremaster being kept moist. The oedema subsides during the night.

The cremaster is then more widely exposed by removing skin and subcutaneous tissue and about 250 ml distilled water is perfused at a manometer pressure of 100 mmHg. This is followed by the stain, usually Mayer's haemalum, at the same

pressure. It is important to begin staining before oedema returns; in oedematous tissue, perfused stain is liable to be restricted to the arterial side with little penetration of the capillaries and veins. Staining is watched under the microscope and when judged adequate, usually after about ¾ h, the vessels are washed out with distilled water. The area of cremaster required for study is then excised, washed in distilled water, blued in tap water and mounted in glycerine jelly.

Other haematoxylins, Loeffler's methylene blue and Mayer's carmalum have also been used but usually with less satisfactory results. Methylene blue renders conspicuous the mast cells alongside the vessels.

To outline the endothelial cells, after perfusing formaldehyde for an hour and washing out with distilled water, 0·25 % silver nitrate is perfused for 5–10 min. The vessels are again washed out with distilled water and then the colour is developed by perfusing PQ developer (Ilford) 1 in 25 for 10–15 min. Staining is watched under the microscope. Pieces of cremaster are excised, passed through fixative, washed in distilled water and mounted as above.

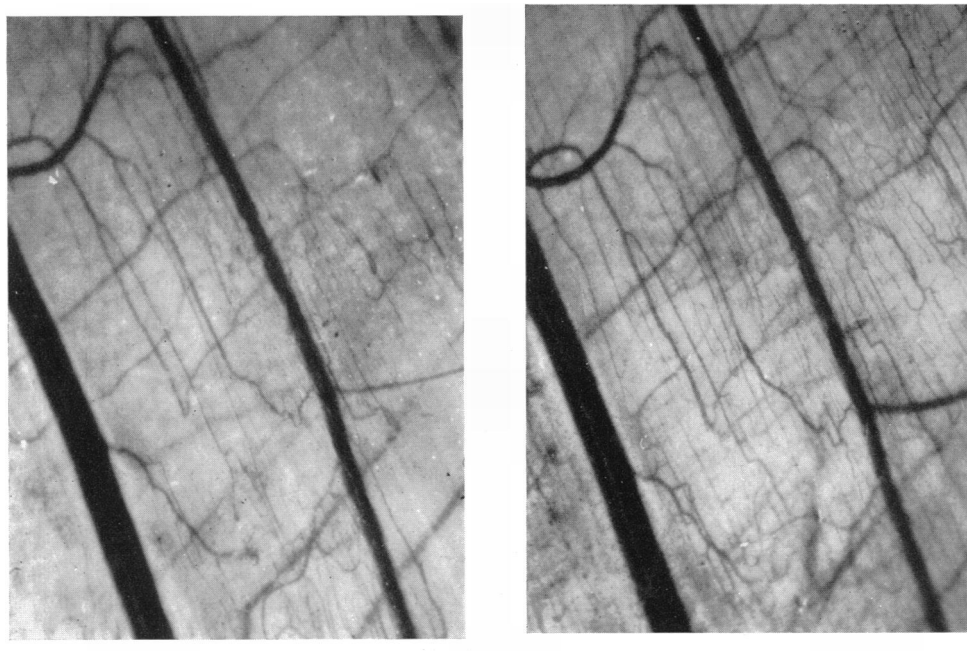
Some portions of the cremaster have been impregnated with silver according to Richardson's (1960) method.

RESULTS

Observations during life

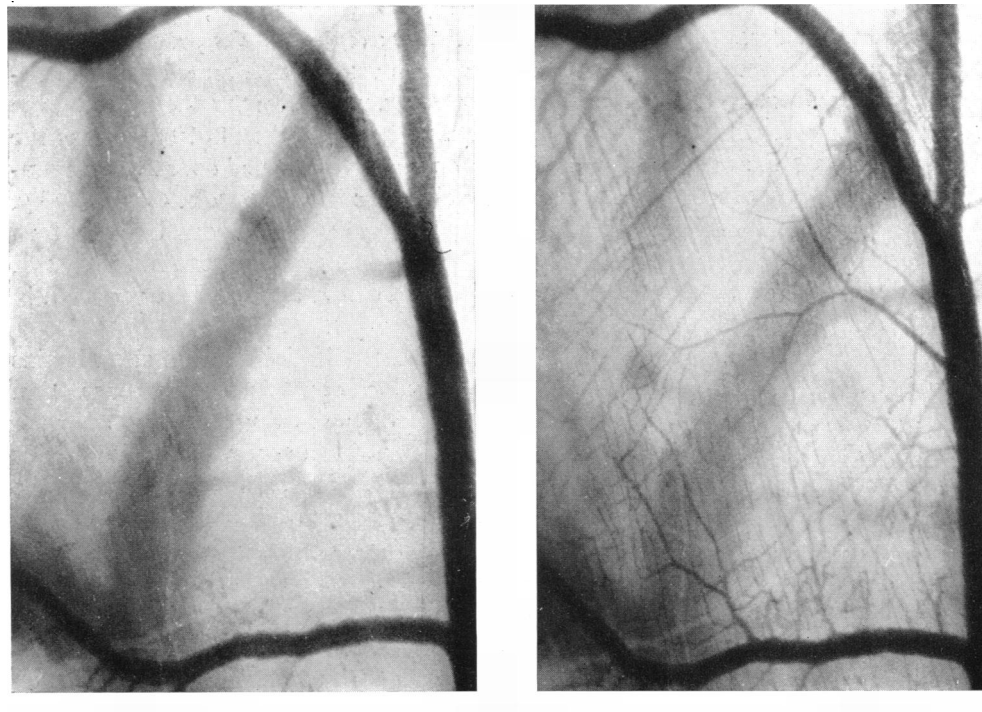
When the muscle is first exposed, the blood vessels are more or less dilated (Grant, 1964). It is usually easy to trace the passage of blood from an arteriole through parallel and intercommunicating capillary channels to a venule. Sometimes one or several channels are more conspicuous by being a little wider than their neighbours and it is usually such channels, if any, that remain open, though narrowed, when the initial dilation subsides. Because of the rapid respiratory movement of the preparation, it is not feasible to measure the diameter of the minute vessels with an eyepiece micrometer, but a measure can be obtained from the photographs. In the dilated state, the capillary blood columns are usually no more, and often less, than 8–10 μm wide; the more conspicuous ones reach 12 μm. Although we have examined over 200 preparations we have not seen wider communicating channels in the muscle. Occasionally, during exposure of the cremaster and before the loose subcutaneous tissue is torn through, a few arteriovenous communications of up to 20 μm are seen in this tissue. They are associated not with the muscle but with the small pads of fat common in this situation.

Provided that the exposed muscle remains quiet, the initial dilation soon begins to subside and the vessels constrict. The degree to which constriction proceeds varies. Figures 1–3 give examples of the vessels seen in both the resting state and after the application of a dilator agent. Sometimes in resting muscle a group of vessels remains conspicuous as in Fig. 1*a*. Here two pairs of longitudinal vessels are seen between the main artery (right) and main vein (left). Circulation remained rapid in these four vessels, which may be regarded as thoroughfare channels for the area. Many more vessels become visible in the dilated state shown in Fig. 1*b*. Frequently, however, in resting muscle almost all the vessels disappear and the area becomes virtually ischaemic as in Fig. 2*a*. The only cremaster vessels visible are branches of a large vein (above and below) while crossing the field are the fainter shadows of



(*a*) (*b*)

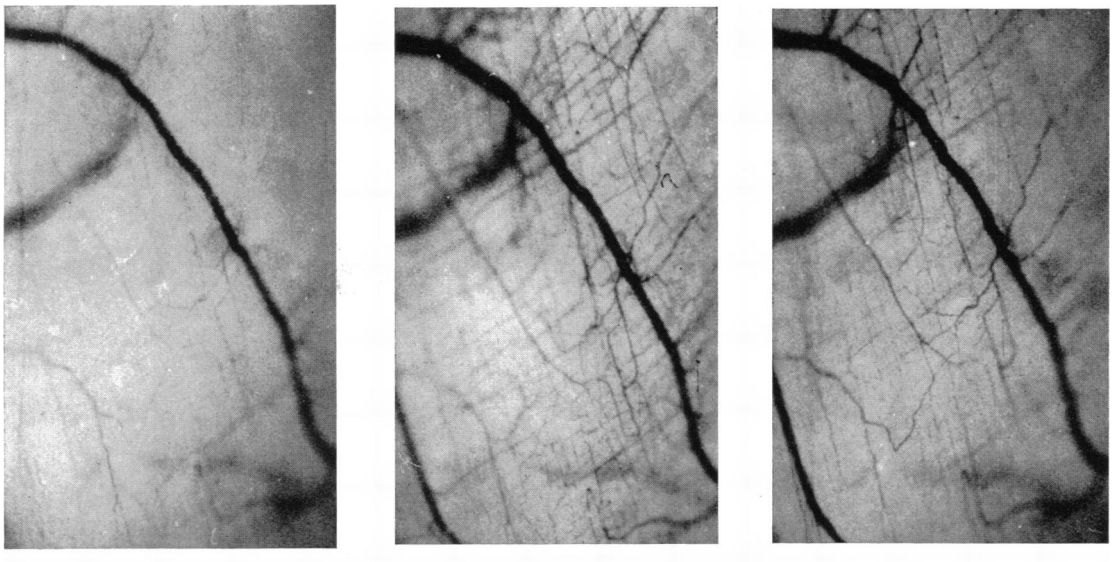
Fig. 1. Living cremaster (×33): *a*, resting state; *b*, after local faradic stimulation.



(*a*) (*b*)

Fig. 2. Living cremaster (×30): *a*, resting state; *b* during irrigation with histamine, 1 μg/ml.

testicular veins beneath the cremaster. Such an ischaemic area may persist for periods of up to an hour but is then replaced by dilation when the muscle begins the uncoordinated, flickering contractions described earlier (Grant, 1964). In the dilated state shown in Fig. 2*b*, a small branching artery enters from the right and two of its terminal branches can be traced through to venules. These two branches had been seen when the muscle was first exposed and they were watched in the expectation that they might remain open to carry on the reduced circulation of the area. But they, too, like the other vessels in the area, disappeared. Sometimes only one or two channels remain open; Fig. 3*a* shows an example which illustrates the vascular pattern at about $\frac{3}{4}$ h after exposure of the muscle. A faintly seen arteriole runs obliquely down from the left and above to fork about the centre of the picture. Each branch can be traced through irregular channels to a venule. In the dilated state these channels are clearly visible, the left branch in Fig. 3*b* and the right in Fig. 3*c* at a slightly lower focus.



(*a*) (*b*) (*c*)

Fig. 3. Living cremaster (×30): *a*, resting state; *b*, during irrigation with acetylcholine 1 μg/ml; *c*, during irrigation with serotonin 0·1 μg/ml.

These examples suffice to illustrate the vascular patterns in the resting state. Although we have made many preparations, we are unable to foretell what the resting pattern would be, nor can we predict what vessels, if any, will remain open, though they are usually the wider ones. But even these, as Fig. 2 shows, may shut down. The general result of our observations of the living muscle is that in the resting state all the vessels except the veins tend to close and often do so. As the vessels constrict, some of the minute ones may remain open and so act as thoroughfare channels.

In the living preparation, the walls of the vessels cannot be seen and, though arteriolar origin and venular termination are obvious, it is impossible to say where the transition between arteriole and capillary or between capillary and venule lies

or what their structures are. For this information we turn to the histological preparations.

In what follows, after a general description of the histological preparations, we deal first with the vessels that closed down in the resting muscle and secondly with those that remained open to act as thoroughfare channels.

Histological observations

(*a*) *General*

When the area that was observed during life is watched while stain is perfused through the fixed preparation, it can be seen that vessels which acted as thoroughfare channels are usually the first to fill with stain from the arteries. As perfusion proceeds, the endothelial nuclei become defined. Soon, colour begins to diffuse patchily around the coloured vessels. The reddish colour of the haemalum entering the arteries changes to bluish in its passage through the capillaries and veins. Figure 4 illustrates the result of 20 min staining. It shows several straight capillary channels darkly outlined; they had been seen to remain open in the resting state during life. The uppermost channel arises from the arteriole on the right and enters a branch of the vein on the left. The endothelial nuclei are visible in the vein wall and are just recognizable in the arteriole to the right and in the branching arteriole in the upper part of the picture. The smooth muscle nuclei of the arterioles are but faintly stained and hardly visible in the figure. The dark patches are due to diffusion of the stain from the vessels. To secure adequate definition of all the vessels and of the smooth muscle nuclei requires staining for at least $\frac{3}{4}$ h.

Preparations perfused with haemalum for about an hour are darkly coloured and show considerable staining of the tissues other than the vessels. Nevertheless, the vessels, and particularly their nuclei, are preferentially stained and are in this way much more clearly displayed than when the piece of cremaster is stained by immersion in the haemalum. The arteries and veins run between the two muscle layers while the capillaries lie between the individual fibres. Microscopically, the various classes of vessels are easily recognized and it is possible, though tedious, to follow the distribution of the stained capillary tubes from artery to vein. It is usually not difficult to identify the vessels seen during life.

Occasionally an arteriole may be found apparently blocked by a small mass of darkly stained granular material. If the injection of heparin is omitted before killing the animal, then many more arterioles may be so blocked. Sometimes a few leucocytes are to be seen within the veins, apparently adhering to their walls. They are more numerous when the cremaster has been exposed for several hours before the animal is killed, and absent when heparin is given and the animal killed before the cremaster is exposed.

With whole mounts in glycerine jelly, there is little shrinkage or distortion of the tissues or alteration of vessel size. Thus, distances measured from point to point in the preparation usually agree well with those measured in the photographs of the living muscle. For example, in a photograph ($\times 50$) two distances at right angles to each other were measured at 75 and 70 mm. In the stained tissue ($\times 50$) the corresponding distances are 75 and 65 mm. Similarly, the diameter of the vessels

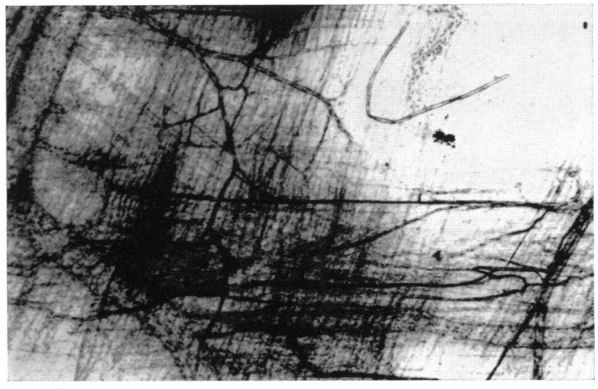


Fig. 4

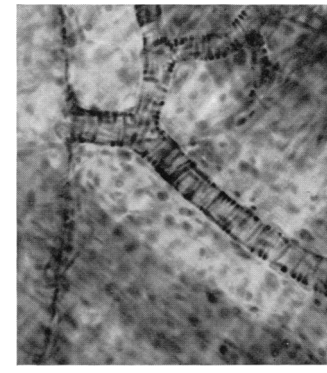


Fig. 5

Fig. 4. Cremaster (×47). Mayer's haemalum. A branching terminal arteriole crosses the right bottom corner; a larger branching vein lies on the left; capillary channels lie between.

Fig. 5. Cremaster (×200). Mayer's carmalum. A small artery runs down the left and gives rise to a branching arteriole.

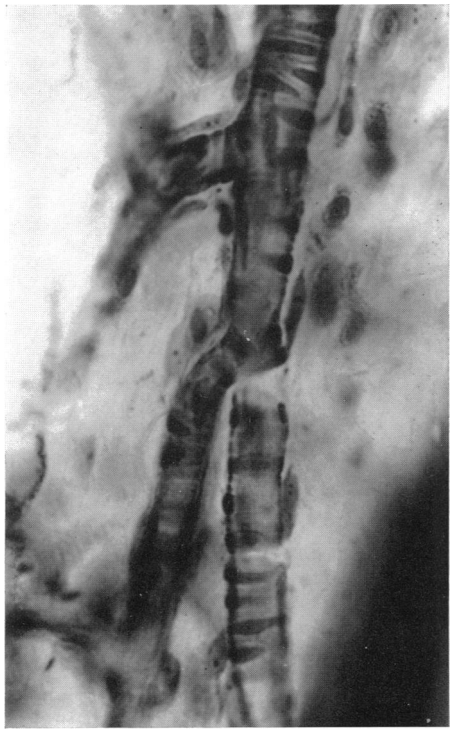


Fig. 6

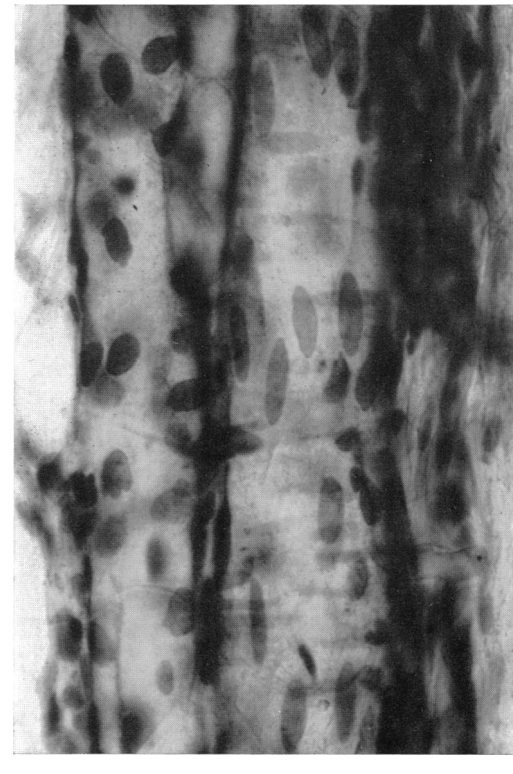


Fig. 7

Fig. 6. Branching terminal arteriole in cremaster (×480). Silver impregnation Richardson's (1960) method.

Fig. 7. Small artery (right) and vein teased from cremaster (×450). Mayer's haemalum.

measured in the photographs (width of red cell column) and in the preparation (internal diameter) show good agreement. Thus, from a photograph the natural widths of two arteries, two veins and two venules were estimated as 80, 40, 90, 90, 40 and 20 μm respectively, while the corresponding diameters in the stained preparation were 80, 40, 93, 95, 41 and 20 μm.

Since there is no standard definition of the various classes of vessels, we define the terms used here for the rat and applied to vessels fixed and stained in the dilated state. An *arteriole* is a vessel with a single layer of smooth muscle and an internal diameter of no more than 30 μm. A *terminal arteriole* is a short vessel of 10–20 μm diameter which breaks up into *precapillary arterioles*. These almost at once lose their smooth muscle cells to become *capillaries*, that is, endothelial vessels of no more, and usually less, than 12 μm internal diameter. The term *venule* is used for an endothelial vessel of internal diameter ranging from 12 to 40 μm. Above 40 μm, smooth muscle cells appear on the vessel wall and the vessel becomes a *vein*.

(*b*) *Vessels that closed down*

We have paid special attention to the smooth muscle cells and to the endothelial cells and their nuclei.

On the *arterioles*, the single layer of smooth muscle cells is continuous to their terminations. The smooth muscle cells lie side by side down to and including the precapillary arteriole. But, as Fig. 5 shows, an appearance of discontinuity is given because not all the smooth muscle nuclei are arranged at the side of the vessel, though the majority are. In looking along the vessel in optical section, the series of rounded knobs presented by the stained nuclei is interrupted by occasional gaps (see also Fig. 9). At the transition to a capillary, the smooth muscle cells of the precapillary arteriole do not disappear gradually but end rather abruptly.

Though there is no thinning out of the smooth muscle cells, the cells themselves become modified in the terminal arteriole. Figure 6 shows that the usual rod-shaped smooth muscle nuclei are replaced by shorter and broader nuclei. In the figure, only the second nucleus from the proximal end of the vessel conforms to the usual rod shape: in addition, the faint outlines of some of the smooth muscle cells can be seen. These, like their nuclei, are also shorter and rounder so that their nuclei are slightly further apart.

In passing from artery to vein, the *endothelial nuclei* change in shape. In the arteries (Fig. 7) the endothelial nuclei are spindle shaped, measuring 20–25 μm long by 2–3 μm wide. They might be mistaken for the nuclei of longitudinal smooth muscle fibres, though they are not so rod-shaped as the transverse ones faintly seen in the figure. The venous nuclei are more rounded, usually measuring 12–14 μm long by 5–6 μm wide, and may even be round. The change from arterial to venous shape takes place in the capillaries. In a long straight vessel such as that shown in Fig. 4, the change in shape occurs about half-way along its length or rather nearer the arterial end. By the change in shape, arterial and venous portions of a capillary may be distinguished. However, it is not always possible to follow the transition along the vessel because in the capillaries (as Fig. 8) many of the endothelial nuclei lie at the side of the vessel and are thus seen edge on. Rounded nuclei are obvious in the venule of Fig. 8.

Perfusion with silver nitrate to outline the endothelial cells shows that the differ-

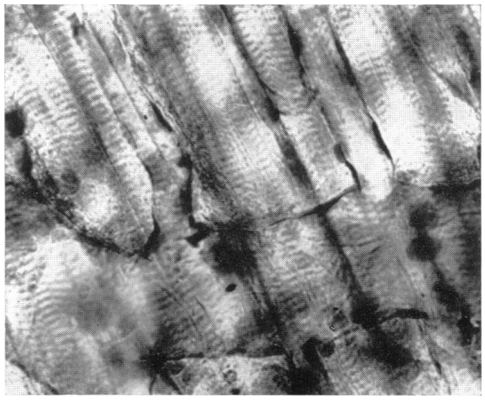


Fig. 8

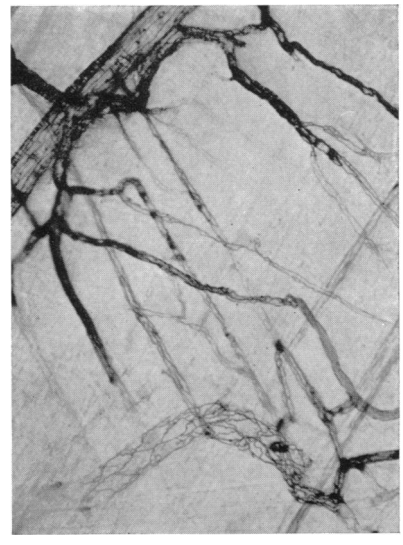


Fig 9

Fig. 8. Venous ends of capillaries opening into a collecting venule (×400). Mayer's haemalum.
Fig. 9. Endothelial cells of cremaster vessels outlined by silver nitrate perfusion (×110). An arteriole crosses the top left corner and gives rise to a branching arteriole and capillaries which join the collecting venule towards the lower part of the figure.

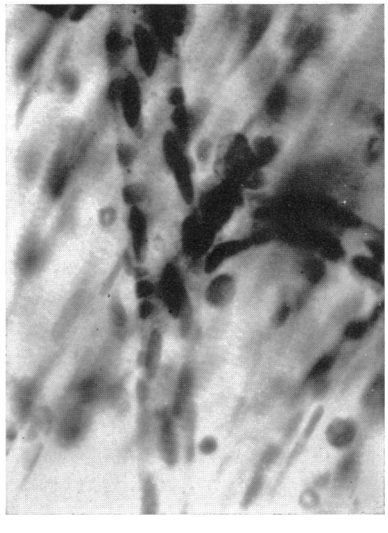


Fig. 10

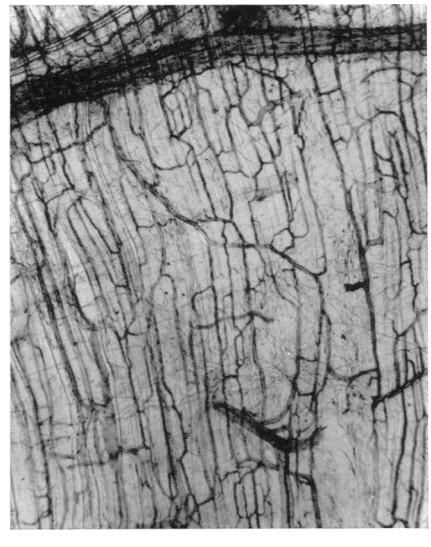


Fig. 11

Fig. 10. Terminal arteriole in cremaster (×400). Mayer's haemalum.
Fig. 11. Vessels in a single layer of cremaster (×38). Ehrlich's acid haematoxylin. A small artery runs across the top and gives rise to an arteriole which breaks up into capillaries. A short portion of a collecting venule lies towards the bottom.

ence in the nuclei is associated with a difference in the shape of the cells. As Fig. 9 illustrates, the endothelial cells of the arterioles and capillaries are long and narrow, while those of the venule are wider and shorter.

(*c*) *Thoroughfare channels*

In twenty-two cremasters we have examined the histology of vessels that had been seen to act as thoroughfare channels during life. We have not found in these any departure from the normal structure. In particular, there is no thinning or discontinuity of the smooth muscle of the arteriole and terminal arteriole leading to a thoroughfare channel; in the capillary part of the channel, the endothelial nuclei change in shape in the usual manner. For example, Fig. 10 shows a terminal arteriole entering from above and dividing into two precapillary arterioles. The vertical precapillary, the direct continuation of the terminal arteriole, forms the beginning of a capillary thoroughfare channel. The smooth muscle nuclei continue uninterruptedly, though with the impression of gaps. They cease towards the lower end of the figure just before the vessel passes out of focus. The endothelial nuclei of both arteriole and capillary are spindle shaped. Moreover, we have been unable to recognize a structural unit with its central arteriolar and venular channel such as is described by Zweifach & Metz (1955). The only commonly recurring unit in the cremaster vessels is the arteriole with its terminal and capillary vessels. In none does the internal diameter of the capillary channel exceed 12 μm. Figure 11 shows the capillary distribution in one muscle layer of an arteriole which arises from the small artery at the top. The only central channel is the ateriole itself; its capillary branches are not reconstituted into a common collecting venule but are diffusely distributed to join with the capillaries from neighbouring arterioles.

DISCUSSION

According to Chambers & Zweifach (1944, 1947), the blood flow through inactive skeletal muscle is restricted to preferential channels which they term thoroughfare or A-V channels. So far as we can gather, the description of these vessels, given for the mesenteric circulation, applies also to that of skeletal muscle and is as follows. The proximal portions of the thoroughfare channels, the *metarterioles*, are relatively straight, one to several hundred μm long, 10–20 μm in diameter when fully dilated and encircled with a single layer of discontinuously arranged but typical muscle cells. Beyond the metarteriole, the muscle cells progressively disappear and the channel, as the *A-V capillary*, continues, until it passes into a venule. The metarterioles arise from terminal arterioles which have the typical arterial structure with a single layer of muscle fibres. Their diameters when fully dilated are not stated but when partially contracted are 20–30 μm. The metarterioles give rise to capillaries. The junctional segment leaving the metarteriole is the precapillary, surrounded with smooth muscle cells, the *precapillary sphincter*.

It seems that this description is derived from observation of the living tissues and is unsupported by histological evidence. Chambers & Zweifach (1944) stated that in the living mesenteric vessels the muscle cells are barely visible, though their nuclei become prominent when the vessel undergoes contraction. In the cremaster preparation, the vessel walls cannot ordinarily be seen. When an arteriole is strongly con-

stricted, its wall is just visible as a refractile streak, but the muscle cells and their nuclei are invisible. As has been pointed out (Grant, 1964), such details as metarterioles or precapillary sphincters cannot be made out, and it was for this reason that we sought histological evidence.

As shown above, the sizes of the vessels in the fixed cremaster correspond well with those of the dilated living vessels. From the description of the living metarterioles given by Chambers & Zweifach (1944) their vessels correspond in diameter and in situation in the vascular tree with our terminal arterioles. But the low power photograph (Zweifach & Metz, 1955) of the blood vessels in rat skeletal muscle (? spinotrapezius), suggests that the term metarteriole includes considerably larger vessels equal in diameter with our arterioles or even larger.

As has been shown, we have not found a discontinuous arrangement or a progressive disappearance of the smooth muscle cells in any arteriole, terminal or precapillary, though an impression of discontinuity may arise from the grouping of the muscle nuclei at the side of the vessel. Further, we have been unable to recognize a functional unit like the 'thoroughfare channel' of Zweifach & Metz (1955). The vessels that remain open are usually slightly wider than their neighbours and so presumably offer an easier passage for blood from arteriole to venule. Apart from this, these vessels do not differ histologically from the other vessels of the cremaster. Even these wider vessels may disappear in the constriction of the resting state.

Zweifach & Metz (1955) stated that in the region of the free edge of the muscle, the metarterioles can be traced directly back to be reconstituted into venous channels. This suggests communications wider than capillaries between arteries and veins. We have mentioned A-V channels of up to 20 μm diameter occasionally seen in the loose subcutaneous tissue surrounding the cremaster which are associated with the fat pads of the region. Algire & Merwin (1955) found on rare occasions a direct connexion between an artery and vein in the panniculus carnosus of mice, but considered that these were not significant in the general circulatory pattern of this layer of striated muscle. Jaya (1963) reported typical arteriovenous anastomoses in serial sections cut from the hind-limb muscles of the rat, either straight or of the glomus type. His brief description and illustrations, however, are not convincing. Sadasivan & Rao (1964) failed to find true arteriovenous anastomoses in rat skeletal muscle and we have not seen them in our cremaster preparation. Barlow, Haigh & Walder (1958) using a variety of methods could not find arteriovenous anastomoses in cat or rabbit skeletal muscle. It is perhaps as yet unjustified to conclude that arteriovenous anastomoses are absent from skeletal muscle in general; they might be restricted to special muscles just as they are restricted to special regions in the skin.

The widening of the smooth muscle cells and their nuclei in the terminal and precapillary arterioles is in accord with Zimmermann's (1923) findings in human material. He remarked that as the arteriole passes into the precapillary vessel, the smooth muscle fibres become modified, widening in the direction of the vessel. Further, Vimtrup's (1923) fig. 2 of an arteriole from the subpapillary plexus of human skin shows the nuclei of the smooth muscle cells as ovoid or round.

Attention has been drawn to the difference in shape of the endothelial cells and their nuclei in the arteries and the veins and the transition taking place in the capillaries. These differences are not confined to the rat. We have re-examined the pre-

parations (intravascular fixation and staining) of rabbit ear perichondrium and human skin made by Grant (1929–31) and Grant & Bland (1929–31). In these also, the long arterial nuclei contrast with the rounder venous ones. The difference is also apparent in tissue fixed without intravascular pressure. Zimmermann (1923) remarked of his fig. 104, that the vessel could be recognized as an artery by the few, narrow and long endothelial nuclei; his fig. 131 shows ovoid endothelial nuclei in a venule. Vimtrup's (1923) figs. 2, 4 and 5 show long, narrow endothelial nuclei in an arteriole, rather round ones in a capillary and ovoid nuclei in a venule. It has been suggested that the shape of endothelial cells varies with the degree of stretching of the preparation (McGovern, 1955); while this might account for the differences in shape in arteries and veins it can hardly explain the change in the capillaries. So far as we know there is no more supporting tissue around the arterial than the venous part of a capillary and it seems to us unlikely that during fixation intravascular pressure would be greater in the distal than in the proximal part of a capillary. But even if the difference is only one of stretching, it may possibly be of functional significance. It has been seen that in our fixed preparations the diameters of the vessels agree well with those seen in the same vessels when dilated during life. We might infer, therefore that, during life, venous endothelium is more stretched than arterial and further, that this might render venous endothelium the more permeable. In this connexion it is of interest that Majno, Palade & Schoefl (1961) showed that in the rat cremaster the endothelial leaks induced by histamine and serotonin are found on the venous side of the circulation, with maximum effect on vessels of 20–30 μm diameter.

The general conclusion to be drawn from these observations is that when skeletal muscle, as exemplified by the rat cremaster, comes to rest, its blood vessels tend to close and often do so, rendering the muscle virtually ischaemic. In this we agree with Zweifach & Metz (1955). Some minute vessels may remain open. These, however, do not correspond with the 'thoroughfare' or A-V channels of Zweifach & Metz (1955); they are often a little wider than their neighbours but are histologically indistinguishable from them, and they do not seem to be vessels specially set apart to carry on the reduced circulation of resting muscle. Why they remain open is unknown. We cannot, however, entirely exclude the factor of injury, for it is known that injury dilates the vessels (Grant, 1964). Injury might arise during the initial exposure or from the subsequent irrigation; but such injury seems unlikely to be responsible for the vessels remaining open, first, because surrounding channels, presumably equally damaged, close down, and secondly because they and the surrounding vessels respond to drugs in physiologic concentration. The finding of leucocytes, however, adhering to the walls of the veins in some of the histological preparations suggests an early inflammatory response to injury.

SUMMARY

1. The blood vessels of the rat cremaster muscle have been studied with special reference to the thoroughfare channels thought to carry on the reduced circulation of resting muscle.

2. The vessels were examined by direct microscopic observation during life and in stained preparations after death.

3. The vessels of resting muscle tend to close and often do so, rendering the muscle virtually ischaemic.

4. Some minute vessels may remain open. These are not histologically distinguishable from their neighbours that close down, though they may be a little wider. They can hardly be regarded as vessels specially set apart to carry on the circulation of resting muscle. They do not correspond with the thoroughfare channels of Zweifach & Metz (1955).

5. Why some vessels remain open is unknown, though the factor of injury cannot be excluded.

6. Attention is drawn to the change in shape of the endothelial cells and their nuclei in passing from artery to vein and to the modification of the smooth muscle cells in the terminal arteriole.

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