# "On the Minute Structure of Striped Muscle, with Special Reference to a New Method of Investigation, by means of 'Impressions' stamped in Collodion."

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[Plate 6.]

## *Historical.*

Curiously enough many of the early microscopists—Schwann for instance—recognised that the fibrils of a muscle are not simply threads of uniform thickness, like those of connective tissue: they were able to demonstrate their varicose character, even with the imperfect lenses at their command. They concluded—of course without any experimental proof—that the cross striping of the fibrils, and, therefore, of the fibres themselves, was an optical expression of such varicosity.

But Bowman, while believing apparently that the striping was optical, and comparing the muscle fibril to a beaded rod of glass, succeeded in breaking up the fibrils into little segments.

According to his view, these "sarcous elements," as he termed them, joined end to end by cement, constituted a muscle fibril. He further believed that each sarcous element coincided in position with one of the alternating stripes, the other kind of stripe corresponding with the position of the cement joining the segments together.

But no sooner had histologists begun to associate the cross-striping with structural differences along the fibrils, than their varicosity was almost entirely lost sight of, and every new stripe (and many were discovered by Dobie, Hensen, and others) was gratuitously assumed to mark the position of some new structure.

There was, however, much to excuse what might at first sight appear to have been a great want of critical acumen, for the application of staining reagents appeared to bring out alternating differences of structure along the fibre. Thus, with logwood or picrocarmine or eosine, the clear stripe (isotropic bands), the dark stripe (Querscheibe; disque épais), the band of Hensen (Mittelscheibe; disque médian), and Dobie's line (Querwand; strie mince), all appear to take on the stain in different degrees, so much so that, in specimens successfully prepared, some stripes appear deeply stained, others hardly at all.

Then, again, Brücke and other investigators demonstrated that the fibrils consist of alternating parts, some of which appear to be doubly, and others singly, refracting.

Overwhelmed by what appeared to be such a mass of evidence, the

histologist of ten or twenty years ago felt bound to assume that the fibril was a very complicated structure, and he never doubted that a muscular fibril consists of a series of alternating and recurring structures. It then became his duty to find out what these structures really might be, and what part they play during a muscular contraction.

The lines of Dobie are often seen as narrow dark bands, which were believed to be membranes (Querwand), and it was held that these membranes separated up the fibrils into little boxes (Muskelkästchen), so that a fibril consists, according to these authorities, of a series of little boxes, joined end to end, containing the substances whose position was marked by the other stripes. Certain of these stripes (the dim ones) were considered to mark the position of solid or relatively more solid substances, and the other stripes (the clear ones) to consist of fluid or relatively less solid substances. The appearance of the stripes, the staining, and their action on polarised light gave, at any rate, some colour to this hypothesis, for the dim stripes appear to have more substance than the clear stripes; they appear to stain with reagents, and to doubly refract light, which latter property is certainly seen in *some* solids. The light stripes, on the other hand, appear deficient in substance and solidity, they stain less readily, and they simply refract light (a property common to all liquids, and some solids).

The Muskelkästchen hypothesis seemed, therefore, feasible enough, and having under their microscopes little boxes containing more solid and less solid parts in alternating layers, Krause, Merkel, Engelmann, and others, sought to explain, each in his own way, the most obvious phenomenon of contractility, namely, the shortening and thickening of the contractile tissue, as being due to the interaction of these structures.

Histologists are accustomed to observe osmotic changes, the swelling up and the shrinking of red blood-corpuscles, for instance, and to see the resulting alterations of form. Under these circumstances it was not unnatural for them to suppose that during contraction the more solid parts of the Muskelkästchen imbibed fluid from the less solid parts, in such a way as to alter the shape of the muscle box, making it shorter and thicker, and causing, in consequence, the whole fibre to change in the same way.

In apparent support of this theory, the stripes in the muscle boxes change their relative thickness, and alter in appearance, in the manner so carefully described by these observers. There seems, in fact, only one objection to this osmotic theory which would at once present itself to the eye of the critical observer; it is the time taken by the process, for osmotic changes are slow in their very nature, and the muscles of an insect's wing can contract over a hundred times a second.

*Personal Observations before the Year* 1880.

More than ten years ago it was my duty, as a young teacher, to make myself familiar with the current literature bearing upon the structure and function of muscular tissue. Even then the number of publications was very great, and I can now recall the despair with which I tried to get a grasp of a subject about which no two observers could be found to agree. While endeavouring to verify some of the statements I had read, I found out for myself that the fibrils are in reality *varicose* threads of tissue, presenting alternate swellings and constrictions of their substance. At once the conviction forced itself upon my mind that the striping might after all be an optical expression of the *form* of the fibrils, and have nothing whatever to do with their internal structure; and it was not until my results were in manuscript form, and ready for publication, that I got access to the older and almost forgotten literature in which I found the same views freely expressed, although without any attempt at their proof.

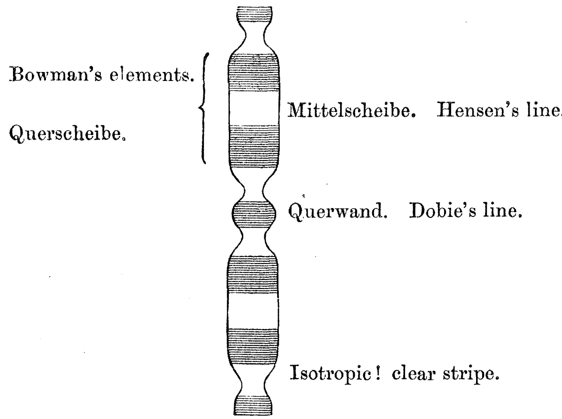
When I had made certain that, both in the fresh and in the prepared muscle, the fibrils are invariably varicose, then I felt that the position of the subject was as follows. Such fibrils are bound to be cross-striped like all other objects of similar shape, viewed by transmitted light. It may be that the cross-striping observed is due to the varicosity alone, or to the varicosity and to co-existing structural differences as well; and, under these circumstances, before we are in a position to take any further step in an investigation into the nature of the muscular fibre, it is imperative to eliminate the appearances due alone to varicosity.

My first endeavour was to ascertain whether there are any stripes that do not correspond in their position with inequalities in the thickness of the muscular fibrils.

Of course in many cases it is difficult, especially if the fibrils or fibres are somewhat distorted, to make out the border clearly; but in good specimens, in a suitable position for study, I found that the striping, both in the contracted and uncontracted fibre, corresponds invariably to either thickenings or constrictions of the fibrillar substance, and in this investigation the muscular tissue of many representative species, both Vertebrates and Invertebrates, was examined. The broad dim stripe occupies the position of a thick bulging part of the fibre, and Hensen's stripe, when present, corresponds to the position of a shallow depression in its centre. The clear stripe lies in the constrictions of the fibril and Dobie's line corresponds with a tiny swelling in its centre.

In addition, the stripes can be reversed by altering the focus, just as is the case with a little varicose glass thread, the scale of a *Lepisma*, or the shadow in the centre of a red blood-corpuscle; indeed Bowman

FIG. 1.




Part of a muscular fibril is represented in this figure, and it will be noticed that the striping of the fibril corresponds with the position of inequalities in its thickness.

actually described the striping in the reverse focus to that generally adopted, calling "clear" what we speak of as the "dim" band.

A very simple method of determining exactly what part varicosity plays in the production of the cross-striping then suggested itself to my mind; it was to immerse the fibrils in a fluid having the same refractive index. Under these circumstances it is obvious that these stripes, which are due to varicosity alone, will disappear, but the striping will become even more marked if there are alternating structures along the fibre possessed of different refractive indices. At Professor P. G. Tait's suggestion, I placed the fibres in a mixture of alcohol and oil of cassia, varying the proportions until I approximated to the refractive index of the fibrils. The striping never entirely disappeared, but it grew fainter and fainter, and I am inclined to explain the partial failure of this experiment on the grounds that, unlike a glass rod, the muscle fibres imbibed the medium in which they were embedded. As a result of this slow imbibition, the refractive index of the fibres would be constantly altering, and it would be a matter of the greatest difficulty to make it exactly the same as that of the surrounding medium. In addition to this, coagulation would be almost certain to take place within the fibrils, destroying their optical uniformity.

While looking upon the results I had obtained as valuable but not conclusive evidence, I sought to solve the problem in another way. I took the living muscle of a Crab or Fly, and, while examining it under the microscope, I pressed down the cover-glass with a

needle. Under these circumstances, those fibres which were pressed upon lost their cross-stripes, and looked extremely like connective tissue. Of course it might be urged that the fibres were by this pressure entirely disorganised, and no conclusions can legitimately be drawn from the experiment, but to this it can be replied that if there really are little bands of tissue so clearly distinguishable from each other, as those who hold the *Muskelkästchen* hypothesis believe, these or their traces should be found scattered about throughout the preparation. In point of fact, as you press upon the cover-glass the stripings gradually disappear with increased pressure, and in the ill-defined fibrillated structure that remains there are no traces of the broken *Muskelkästchen.* And finally, if more proof is wanted, it is possible by means of a screw, which raises or lowers the cover-glass, first to press upon the fibres and cause the striping to disappear, and then on raising the cover-glass to cause them to reappear once more. We can only explain this result on the assumption that the varicose fibrils are flattened out, and that the striping caused by their varicosity disappears in consequence.

There were, however, three important facts which had to be thoroughly accounted for, before it could be affirmed that the fibrils do not consist of the alternating structures supposed to exist; these facts were the effect of cleavage, of staining and the action of polarised light. The muscle fibrils can be broken across into the sarcous elements described by Bowman; but a careful study of the question soon convinced me that the cleavage *is always across the thinnest parts of the fibrils*, taking place in the substance of the clear stripe. If Dobie's line is at all marked, the cleavage takes place near the little swelling which corresponds to it, and through the substance of the clear stripe. Reference to fig. 1 will at once show that here we have to deal with the thinnest part of the fibrils, and it is therefore begging the question to assume anything over and above this mechanical reason for the cleavage, for every varicose rod will break across at its thinnest part. The phenomenon of transverse cleavage cannot therefore be taken in itself as an argument in favour of structural differences along the fibrils.

The appearances seen in stained preparations can also, I pointed out, be satisfactorily explained on the varicosity hypothesis. We find that whatever else is employed, and at whatever focus you adopt in your examination, those stripes which in the unstained preparations appear dim also appear to take on the stain, while those stripes which appear clear and bright are unaffected by it. In fact, the difference in colour is entirely a question of "saturation," for whenever there is a flood of light, as in a clear stripe, the colour of the fibre at that part becomes unsaturated by it. It is easy to convince oneself practically of this fact by the examination of varicose threads of faintly coloured

glass *in a ray of parallel light.* Rods of faintly tinted glass of the same shape as the muscle fibres, having tiny globules—Dobie's lines—and broader swellings for the dim stripes, when examined under the microscope, or in the field of a lantern, give as strong colour differentiations as any muscle fibres, the constrictions coming out quite colourless, while the dim band and Dobie's line are sharply brought out by their deepened colour.*

One of the chief faults of which I was guilty when publishing these results in 1880 was that I did not sufficiently lay stress on the appearances presented by a coloured or colourless varicose thread of glass when placed in the path of a parallel ray of light. It is quite different from the same thread when examined in diffuse daylight, for in the latter case a hundred images fall upon the retina at the same time, and the striping and colour differentiations are confused. One can see little appearance of striping, and if the glass is coloured it may appear very much of the same tint; place it in a lantern, or even lay it down on a piece of white paper, and the picture is quite different. As one is accustomed to view objects in diffuse daylight, one is not prepared to interpret correctly the character of such an object when viewed through a microscope: the clear well-defined bands and colour differentiations of a muscle fibril are not the appearances of a varicose thread as seen in diffuse daylight, but they are those of a similar fibre observed in parallel light when practically a single image falls upon the retina.

Lastly, we come to the action of polarised light, and here at once the phenomena by no means prove structural differentiation along the fibre. There are many questions which lead to complication. We have the varicosity of the fibril, which will alter the path of the polarised beam and produce apparent differences along the fibre when there may be in reality none at all. Then we have as a complication the interfibrillar substance, which is simply refracting, and which is chiefly lodged in the neighbourhood of the clear stripe. I was not prepared to say, under these circumstances, what is the action of polarised light on the fibrils, nor do I wish to commit myself now: it is sufficient to say that, even if we grant that alternating singly isotropous and doubly refracting anisotropic bands exist along a fibril, it does not follow that these are bands of more solid and less solid material: the whole difference may be due to molecular tension. A fibre of such a shape, as was pointed out to me both by Professor Stokes and by Professor P. G. Tait, is almost bound to possess alternating parts in different conditions of molecular tension, and give the familiar appearances when examined by polarised light.

* In doing this experiment, only faintly tinted glass must be used, and, as this is difficult to obtain, I generally use hollow varicose tubes of white glass filled with coloured fluid.

A paper containing the above results was presented to the Royal Society of London by my kind friend Professor E. Klein, and was printed in the 'Proceedings' of 1880, and in the 'Quarterly Journal of Microscopical Science,' 1881, and in this paper I ventured to assert that I had been able to explain the appearances generally considered to indicate structural differences in the course of the fibrils as being due to the varicosity of the fibrils themselves. I further stated that of course structural differences might exist, but that the proof of their existence was not as yet forthcoming.

My views were received in many quarters with kind consideration, but they were only very partially accepted. For my own part, as soon as I had published them I resolved not to think about the subject again for some years, when, with more matured experience, I might return to its consideration and picking up the threads that I had dropped unravel them with a more skilful hand.

### *Recent Investigations with the Collodion Impressions.*

Last winter (1889–90) an idea occurred to me which led once more to my examination of the subject. It struck me that if I could "stamp" some soft transparent solid with muscle fibres it might be possible to obtain impressions of the fibres on the soft material. If these impressions had smooth unstriped depressions corresponding to the fibres, this would indicate that the striping was caused by structural differences *within* the fibrils; if, however, the impressions were striated, this could only be explained on the ground that the striation of the "stamp"—the muscle—was caused by the *form* of the fibrils, which form and which striation were transferred to the soft material in the process of stamping.

It seemed improbable that I should succeed in getting faithful impressions of such microscopic objects, yet I felt that it would be well worth while making the attempt, for the results if obtained would be most conclusive. I experimented with every substance that I could think of, using wax of various kinds, glass, gelatins, glycerin jelly, transparent soaps, &c. Once or twice I thought that I had obtained very partial success, but my difficulties were great, for whenever I hit upon a substance like gelatin, for instance, which would set in intimate contact with the fibrils, it invariably came away with them when they were removed. I worked at the subject for months, trying every expedient which suggested itself to me, and in July, 1890, I at last succeeded beyond my most sanguine anticipations.

It occurred to me that perhaps collodion might be of service, for a thin layer dries quickly and forms a beautifully smooth transparent film. I accordingly prepared a film by allowing a drop of collodion to fall upon a slide, and tilting the slide so that it flowed over it in

a layer of uniform thickness. When still somewhat moist I pressed against the film some roughly teased muscle fibres held on my finger tip. They came away quite readily when the finger was removed, leaving little "ruts" in the collodion obvious to the unaided eye. On examining these ruts with the microscope I found what I at first thought were actual muscular fibres still adhering to the collodion film, showing the fibrils and every detail of the cross striping with remarkable clearness. The ruts contained, however, no trace of muscular tissue when examined by the naked eye, for the slightest trace of muscle is at once recognised by its opacity. On looking at the specimen a few minutes afterwards, what was my surprise to find that all the appearances I had just seen had completely vanished, the ruts had disappeared, and the collodion film was flat and smooth.

The explanation was very soon found, and no doubt remained that what I had at first actually mistaken for muscular fibres were in reality their "impressions," their subsequent disappearance being due to the contraction of the film, as it dried, pulling out every inequality in its surface.

It is very instructive to watch one of these collodion impressions; at first clearly cut, with every stripe sharply defined, they gradually fade, and perhaps in five or ten minutes they disappear entirely. Sometimes a portion of a fibre really remains sticking to the collodion; it is at once recognised by its great opacity. What astonished me almost as much as the perfect reproduction in the impression of every cross stripe was the ease with which these impressions can be made. One can hardly fail to obtain them, and at the International Congress in Berlin, while demonstrating the subject to the members of the Physiological and Anatomical Sections, I made over one hundred preparations—few of which were failures. Not only can one stamp with hardened muscle, but the fresh tissue can itself be used. Of course the fresh tissue is soft and does not make so good a stamp, the results are not so striking, but they are quite evident. In making impressions of a fresh muscle one can take a piece of muscle, say from a Rabbit, cut it through in the direction of the fibres, and press the cut edge for a second or so against the collodion film, which must be very soft: one rarely examines the film without getting some trace of an impression upon it.*

If the impressions are examined with a high power, say 600 diameters, the following details can be made out. Each fibril, if a hardened

* One can get impressions of other tissues, bone, tooth, hair, &c. A section of dry bone comes out very well, and one can see in the impression the "set" of the lamellæ, the lacunæ and their canaliculæ, and every detail with marvellous clearness. If a still moist film be pressed against the back of the hand, and then examined, one sees the impressions of the imbricated scales covering the hairs on the back of the hand far clearer than in the original.

preparation be used for stamping, makes its own individual impression in the collodion, which rises between the fibrils in the place of the interfibrillar substance, which has, of course, been removed in the ordinary preparation of the tissue. When the muscle is pulled away the impressions of the individual fibres can readily be made out, and the borders of the little varicose hollows are plainly to be seen; the cross-striping, which can here only be due to the *form* of the impression, is exactly the same as that of the muscle itself. To put it in another way, we have varicose threads of air, within surrounding collodion in the place of varicose threads of muscle surrounded by balsam or Farrant, and varicosity, the only common factor in the two cases, is alone the cause of the striping observed in each. Not only are the broad stripes well marked, but one can see with even greater ease than in the muscle itself the lines of Dobie and of Hensen. In the fresh muscle I have only once or twice seen the outlines of the fibrils with any degree of distinctness, but the stripings are more readily seen; yet one would hardly expect to get such good results from fresh muscle, both on account of its softness and from the fact that the fibrils are covered by sarcolemma. If the collodion be tinted, say with magenta or Bismarck-brown, impressions can be made in this coloured medium, and these show in beautiful detail the apparent stain differentiations observed in muscle. The broad dim stripe comes out red and appears like a solid, well-defined band, and the clear stripe in successful preparations appears by contrast devoid of colour.

It will be seen from the above experiments that, as the stripings are all optical effects of varicosity, the very foundations of the Muskelkästchen hypothesis are removed, and we now come to the consideration of the phenomena of *contraction.*

*An "impression" of a muscular fibre shows in every detail the appearances characteristic of the muscle used to stamp it, in whatever state of contraction or relaxation it may happen to be.*—If a piece of muscular tissue, hardened in alcohol in the extended position, be examined under the microscope and its details studied, and if an impression of it be then made, the impression will show the same details that it shows. The same holds good for the contracted or semi-contracted fibre. Photograph I was very kindly taken for me by my friend Dr. Carrington Purvis, and shows the appearances presented by a Crab's muscle in a state of extension. The little varicose fibrils are seen separated by little varicose dark lines, the latter being the optical sections of the interfibrillar substance.

Photograph II* is taken from an "impression" of a muscle in a

* The photographs of the "impressions" were taken by my friend Dr. Edington, to whose skill and interest I am much indebted. As the "impressions" only last about five or six minutes, and as with ordinary illumination an exposure of

similar condition, and it will be noticed that the appearance is essentially the same, except that the stripes are reversed, the little dots forming Dobie's line and the dim bands coming out bright, and the clear stripe appearing dark; the slightest alteration of the focus would have reversed the photograph and have given the ordinary appearance.

A contracted fibre has quite another appearance, for not only are the cross-stripings much nearer together, but they have changed in character. Without going into detail, at present, it is sufficient to say that alternately dark and light stripes are seen, and that the Dobie's line, so constant a feature in the extended fibre, is no longer to be seen in the contracted condition; the stripes, moreover, have altered in thickness relatively to one another. Now it is needless to again point out that the change in the striping has hitherto been held to indicate changes *within* the fibrils of the nature of osmosis, the stripes being taken to represent actual structures. But if an impression be taken of a muscle killed in contraction it shows every detail of a muscle *in that condition*, as photograph III, taken from a collodion impression, very well indicates. (In this photograph the clear stripes come out clear, and the dark stripes dark, just as in the original muscle, but of course the appearance could be reversed by altering the focussing.)

It follows that when a muscle passes into a condition of contraction the changed appearance is entirely due to a change in its form, and I have frequently stamped muscles which show in the same fibre both the contracted and uncontracted state with the intermediate stages. These intermediate changes come out perfectly in the "impressions," so that one can positively affirm that the striping is due to form, and every change in striping observed during contraction depends upon some change of form too. Of course the imbibition theories of Krause, Merkel, and Engelmann are no longer tenable, since the facts on which their theories were founded have received another explanation. The Muskelkästchen was evolved on the supposition that the cross stripes correspond to membranes and layers of tissue along the fibres, whereas the impressions prove that they are due to variations in the thickness of the fibrils in different parts of their course. The imbibition theories were evolved on the supposition that the changes in the striping observed during contraction are due to alterations in

from ten to fifteen minutes is required, our first attempts were not as successful as might have been desired, and those exhibited in Berlin were decidedly faint and wanting in density. Dr. Edington subsequently, adopting a suggestion of Mr. Forgan, used magnesium light in the place of the ordinary oil lamp, burning about one foot of the thin riband in the optical axis of the apparatus. This exposure, lasting only a few seconds, gave us very beautiful negatives, from which the photogravure plate was taken.

the relative quantities of fluid held by the substances producing the striping. Inasmuch, however, as the changes in the striping are due to changes in *form* of the fibrils, the very foundation of these theories has been removed.

### *The Author's Views as to the Structure of Striped Muscle.*

Before proceeding further I would venture to state what I think we are in a position to affirm respecting the structure of striped muscle. The fibres consist of fibrils generally grouped together in bundles and separated from each other by interfibrillar matter. As the fibrils are varicose, and have a different refractive index from the interfibrillar matter in which they lie, they, in consequence, present the optical striping possessed by all such bodies under similar circumstances, and we have no reason to suppose that this striping has any other interpretation. The fibrils, from whatever point we look upon them, are composite structures, and their varicosity indicates this quite clearly. Each fibril has practically undergone segmentation into a series of tiny particles, although there is no evidence that these are separated from one another by membranes, or any other anatomical structures, and each little bit contracts on its own account so as to thicken and shorten. Although we know absolutely nothing as to what there is *within* each fibril, yet the condition of parts, whatever it may be, is probably the same in every Dobie's line, or in every dim or light stripe. Each light stripe may merely consist of contractile tissue in a different state of tension from that in the position of the dim stripes, and if so, that may partly interpret the polariscopic phenomena, but beyond the fact that a difference exists we are not in a position to make a further affirmation. When we study the change in form which these little segments undergo in passing from the relaxed into the contracted condition we come upon several curious facts, the interpretation of which is at present very difficult. Some muscles, and especially those of some of the lower Vertebrates appear to be very simple in form, and to undergo very simple changes during contraction. I hope to enter into greater detail in a subsequent paper, but in the meanwhile I would simply state that the fibrils seem to be devoid of the tiny swellings which form the line of Dobie. The fibrils, therefore, possess simply alternate swellings (dim stripes) and constrictions (clear stripes). During contraction, the swellings become more marked as the fibrils shorten, the change being represented in fig. 2.

In this case the dark stripes of the contracted fibre are at just the same parts of the fibril as in the relaxed condition. In other muscles in most of the Arthropoda, for instance, the stripes are reversed, as already so well described by the German histologists.

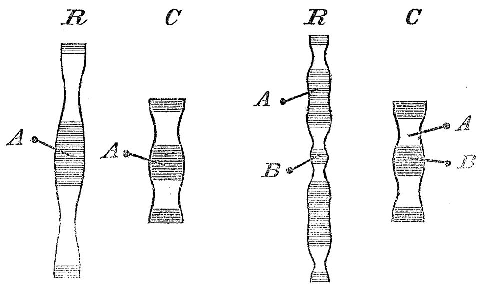


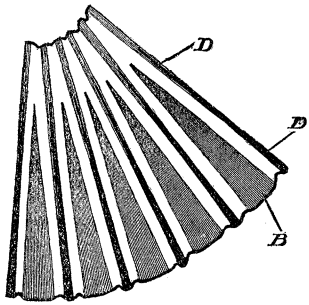

Fig. 2 (*R*) represents a relaxed fibril with a pin, *A*, sticking into the dark stripe. During its contraction (*C*), as the fibril simply shortens and thickens without otherwise changing its shape, the needle *A* is still seen sticking in the dark stripe.

Fig. 3 (*R*) represents a relaxed fibril with a pin, *A*, sticking into the dark stripe, and another pin, *B*, sticking into the swelling in the position of Dobie's line. When contracting (*C*), the fibril is profoundly modified in shape, the pin *A* sticking into the clear stripe, and the pin *B* into the centre of the dark stripe.

The reason is that, during contraction, the fibrils change their shape in such a manner that the parts which previously bulged now become the thinnest part (fig. 3). As the fibrils begin to contract the substance of the clear stripe becomes an eminence instead of a depression, and the little bulging part forming Dobie's lines becomes smoothed out and gradually obliterated. The dark stripe, on the other hand, becomes the constriction in the case of the contracted fibre, and, of course, appears now as a clear band. These points can only clearly be made out by studying all the intermediate conditions between complete contraction and relaxation, and they are best seen in the living muscle fibres on which waves of contraction are still slowly passing; one may see them, too, upon the muscle impressions. I have never happened to make an impression of a fibre showing a series of these intermediate stages in a short piece of a fibre while engaged with Dr. Edington in photographing them, but fig. 4 shows very well the appearance; it is a careful drawing of a Crab's muscle in a state of contraction, but bent at an angle so that the convex side is artificially extended. The Dobie's lines on the extended side are seen gradually to thin away, and gradually disappear on the contracted side, while the surrounding bands which appear as clear depressions gradually become dim elevations.

Of course, this change of form leads to the shortening of the fibrils, but it is at present difficult to say why this reversal of the varicosity should occur; at present, we have to accept it as an unexplained fact.

FIG. 4.



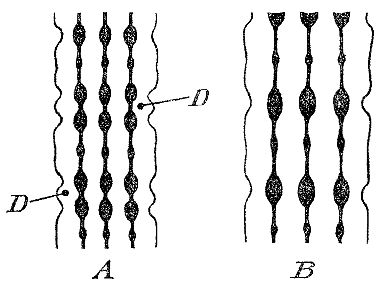

A drawing of a living and contracted Crab's muscle, which has been bent round and artificially extended on its convex lower border. The transitions between the relaxed and contracted parts are well seen. Dobie's lines (*D*) gradually fade away as you pass to the contracted condition, becoming invested by and then replaced by the dark stripe of the contracted condition. The dark stripe of the relaxed part (*B*) fades away, and is replaced by the light stripe in the contracted part.

Many of the German histologists have described a condition observable before the muscle has completely contracted (*Uebergangsstadium*), in which all striping has disappeared, and this Merkel and Engelmann each explains on his own imbibition theory. Now, one can dogmatically affirm that in the greater number of fibres of which the tissue is in one part contracted and another part relaxed, and in which the intermediate stages are plainly visible, as in fig. 4, for instance, not a trace of such a condition is visible; it is therefore not an essentially intermediate phase. I have had the privilege of seeing Professor Engelmann's preparations, and here it is seen, and in my own I *occasionally* come across it; but what I have invariably observed is this, *that it is never seen in a fresh preparation free and unattached to cover-glass or slide.* It frequently happens that fibres become pressed and otherwise fixed, and then it appears that when they shorten, the contracted part pulls upon the still extended portion, and diminishes by so doing the varicosity of the fibrils, and, in consequence, the striping which depends upon it. The effect is exactly the same as that produced by flattening out the fibres by pressing on the cover-glass; in one case the varicosity is diminished or obliterated by a pull in the length of the fibril, in the other case by pressure applied to its sides. One may, at any rate, state that in the vast majority of cases, as the varicosity becomes reversed, the fibrils never become uniform threads of tissue, for, as the dim stripe is flattening out, eventually to form a depression, the clear stripe, with Dobie's line still visible in its centre, is becoming a ridge.

*The Interfibrillar Substance.*

The interfibrillar substance is not usually held to have the property of contractility, and it appears to me that the arguments based on its fancied homology to a cell network recently brought forward, especially in England, can hardly be said to prove the contrary ; I hope to refer to this subject in a subsequent paper, to be devoted to the comparative histology of muscle. From the varicose character of the fibrils, it follows that the interfibrillar substance is of the nature of a matrix or bed of tissue perforated by varicose tubes. It is like a honeycomb minus its transverse partitions, or, better still, like a mitrailleuse ; we have, however, to imagine the walls of the honeycomb of variable thickness, sometimes thicker and sometimes thinner, and the analogy is complete. In optical section, as when we focus a piece of muscle, this interfibrillar honeycomb will appear as in photograph I or fig. 5.

FIG. 5.




Two fibres are represented, *A* and *B*. The interfibrillar substance is strongly represented by the varicose lines; the outlines of the fibrils are faintly represented at the borders of the figures. In *A* the fibrils possess well-marked Dobie's lines; the swellings of the fibrils causing them are seen, *D*. In consequence, the cement matter forming single masses in *B* is in *A* divided into two sets (heads of Schäfer's muscle rods). In *B* Dobie's lines are not seen. In diagram the cross-striping of the fibrils has been omitted for the sake of simplicity.

Here the fibrils are left blank, and the interfibrillar substance is represented by dark varicose lines, the optical sections of the longitudinal walls of the honeycomb. The walls are thick opposite the position of the constrictions of the fibrils which lie within the honeycomb, and thin where the bulgings come. When a Dobie's bulge is present as at *A*, the bulgings, corresponding to the clear stripe, are divided into two (Schäfer's muscle rods) ; but when Dobie's line is not well marked, we have the appearance seen at *B*. Of course, it will be understood that where these thickenings of the honeycomb

occur the fibres are encircled by a thicker band of interfibrillar substance, that the little beads or swellings in the diagrams are merely optical sections of the thicker parts of the honeycomb. These thickened portions, when very strongly differentiated from the fibrils, as by the gold method, may appear like transverse bars running across the fibrils in the region of the clear stripe, and the whole structure has unfortunately been misinterpreted by some observers into a network, the transverse links of which are the encircling and thickened parts of the honeycomb, while the longitudinal threads are the lines really marking the optical section of the honeycomb tubes. If any threads of tissue are to be actually seen, I quite agree with Professor Klein in ascribing them to precipitation within the interfibrillar honeycomb.

*The Physiological Explanation of the Varicosity.*

I may not unreasonably be asked to supply some hypothesis of my own in place of the exploded theories of imbibition, for, if we simply view a muscle fibre as consisting of varicose fibrils, we have a bare morphological fact without any physiological significance. Before doing this, I will venture to clear up one misunderstanding which has arisen concerning the morphological difference between striped and unstriped muscular tissue, although this question will, I hope, be more fully discussed in a subsequent paper.

The unstriated muscle is generally described as a nucleated spindle, presenting fine longitudinal fibrillation, and devoid of a true sarcolemma, while the striped or voluntary fibre is described as a fibrillated thread of contractile tissue, invested by a sarcolemma underneath which numerous nuclei are placed. The heart muscle is generally looked upon as a tissue intermediate between the two. But authors to whom we owe these ideas, have restricted their enquiries to the Vertebrate histology alone. It is necessary to pass into the region of comparative histology, before we can thoroughly comprehend the subject. If we do this, we shall find that there are two chief varieties of fully differentiated muscular tissue. First of all, there is the nucleated spindle devoid of sarcolemma and made up of fibrils cemented together, and we notice that *these spindles may be striped or unstriped, the difference depending upon the rapidity of their contraction.* They are found in most divisions of the animal kingdom; thus, in the adductor muscles of *Cardium*, *Pecten*, *Lima*, rapidly moving Lamellibranchs, we have nucleated and striped spindles; these occur in the heart muscle of the Frog and many other animals, while non-striped spindles are found in parts of the circulating and digestive systems where less active movements are required.

Then again, there is another type of muscular tissue, consisting of

cylindrical threads, sometimes invested by a sarcolemma, and with nuclei within the fibrils, under the sarcolemma or in both of these situations, and we notice that *these threads of tissue are striped or unstriped according to the rapidity of their contraction.* In Vertebrate skeletal muscle, they contract quickly and are striped, and the same applies to the adductor of the *Terebratula* which closes its shell so quickly as sometimes to nip its protruding siphon. In many of the Polychætæ, in many Lamellibranchs, as in *Mytilus*, and in slowly moving Ascidia, the fibres are devoid of striation. We see then that the striping of muscular tissue cannot be said in any way to associate itself with any particular "build" of cell; it may be present in both a spindle and in a cylindrical thread. When a muscular fibre, it may be spindle-shaped or cylindrical in shape, is called upon in the process of evolution to contract very quickly, then it becomes striped, the cause of which is the segmentation of the previously cylindrical fibrils into varicose threads. The Swallow in its rapid flight has quickly to see, and catch the passing fly, and the fibrils of its ciliary muscle, simple threads of uniform thickness in some ancestral form, now become beaded and cross-striped.

Striated muscle may, therefore, be defined as "muscular tissues, the ultimate fibrils of which have become varicose, and this in association with the power of quicker and more active movement."

We can now ask ourselves whether it is not possible to explain this correlation between the segmentation of a muscle and its power of contracting more rapidly, and it will, I think, be seen that a very simple and straightforward explanation can at once be given. The whole subject can be resolved into a question of "mass"; the larger the contractile element is, the longer time will it take to reach its maximal degree of shortening, so that when a fibril segments into a number of much smaller particles, each one contracting and relaxing on its own account, a considerable amount of time will thereby be gained. We have many examples of the influence of bulk, or mass, upon rapidity of contraction in the case of the gross muscles themselves, the larger animals moving relatively slower than the smaller ones, as when the Hare, in spite of its smaller leaps, can nearly keep pace with the Horse, because its leaps are repeated at much shorter intervals. We can now see how, by simple means, a muscle can, during its evolution, contract more quickly, but the fundamental explanation of the phenomena of contraction is still to be found. Whether or not we may ever be able to express muscular contraction in terms of those phenomena which we see in the inorganic world I am not in a position to say, but this we must all be certain of, that this explanation will result rather from a study of the contraction phenomena of the lower and simpler types of con-

Fig. 1.

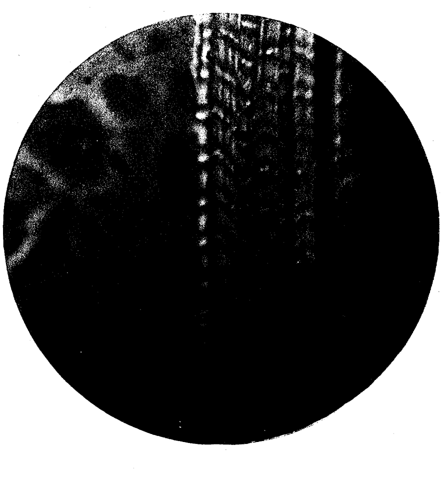


Fig. 2.

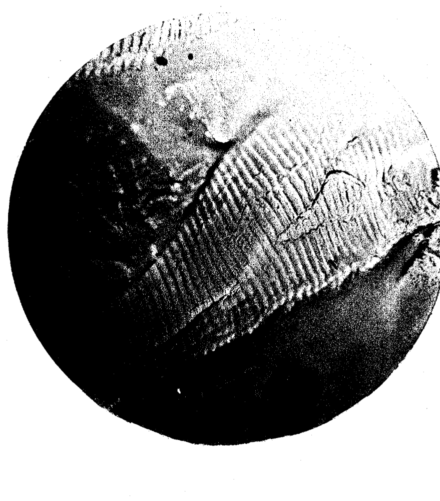


Fig. 3.

tractile tissue than from the highly evolved tissue of the striped muscle.*

## EXPLANATION OF PHOTOGRAPHS. (Plate 6.)

*Photograph I.*

Photograph of the muscle of a Crab in a state of relaxation, and magnified 1000 diameters. Dobie's lines are seen as narrow dark bands running across the fibres, and corresponding to tiny bulgings of the individual fibrils; seen best at the upper edge of the fibre. The clear stripes on either side of Dobie's lines correspond with constrictions in the fibrils, and the dark stripes correspond with broad swellings. The cement substance between the fibrils appears light in colour.

*Photograph II.*

Photograph (700 amplifications) of a moist film of collodion, upon which a piece of relaxed Crab's muscle had been pressed and had then been withdrawn. In this "intaglio" all the appearances of the relaxed Crab's muscle are to be seen, those parts which are dark in Photograph I coming out white in the intaglio. The cement matter and the clear stripes are dark, and the dark stripes and Dobie's lines come out light in colour.

*Photograph III.*

Photograph of a moist film of collodion, upon which a piece of contracted Crab's muscle had been pressed and had then been withdrawn. The striping is that of the contracted fibre in all its detail; the approximation of the cross-stripes to each other and the absence of Dobie's line are points especially to be noted. Owing to the collodion film varying in its thickness, the intaglio is photographed at different focal planes, and the dark stripe, which appears light in the lower part of the photograph, comes out dark in colour at the upper part. The edge of the intaglio is better seen than in Photograph II, and by the aid of a lens one can readily see in the original negative the interfibrillar matter.

I am much indebted to the Cambridge Engraving Company for the excellent manner in which the photographs just described have been reproduced.

* Professor P. G. Tait has recently suggested to me, that, owing to their varicosity, the fibrils will be able, as it were, to get a better "grip" of the interfibrillar matter, so that during contraction or relaxation the muscle will be able more effectually to move as a whole.

FIG 2.

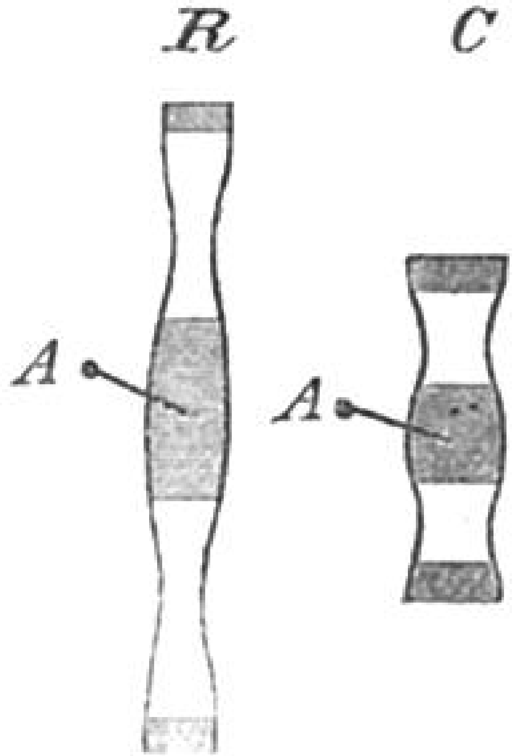



FIG. 3.

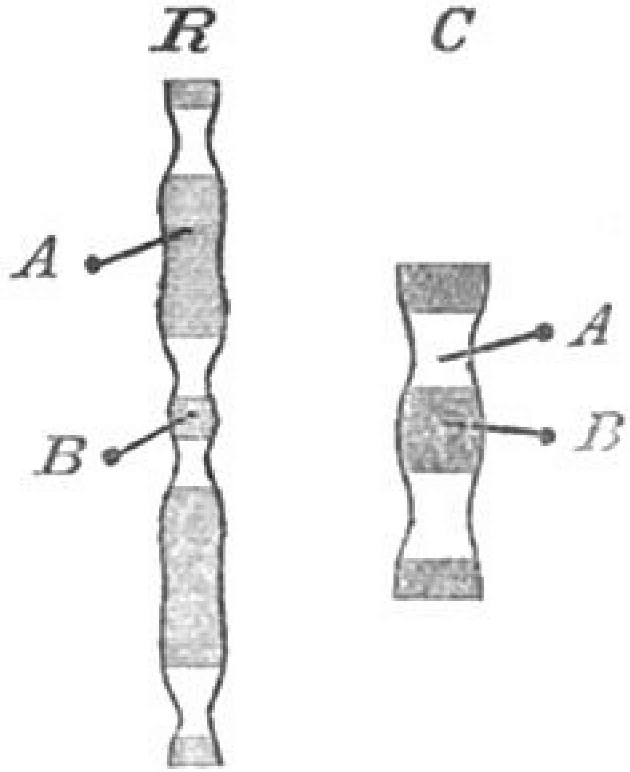
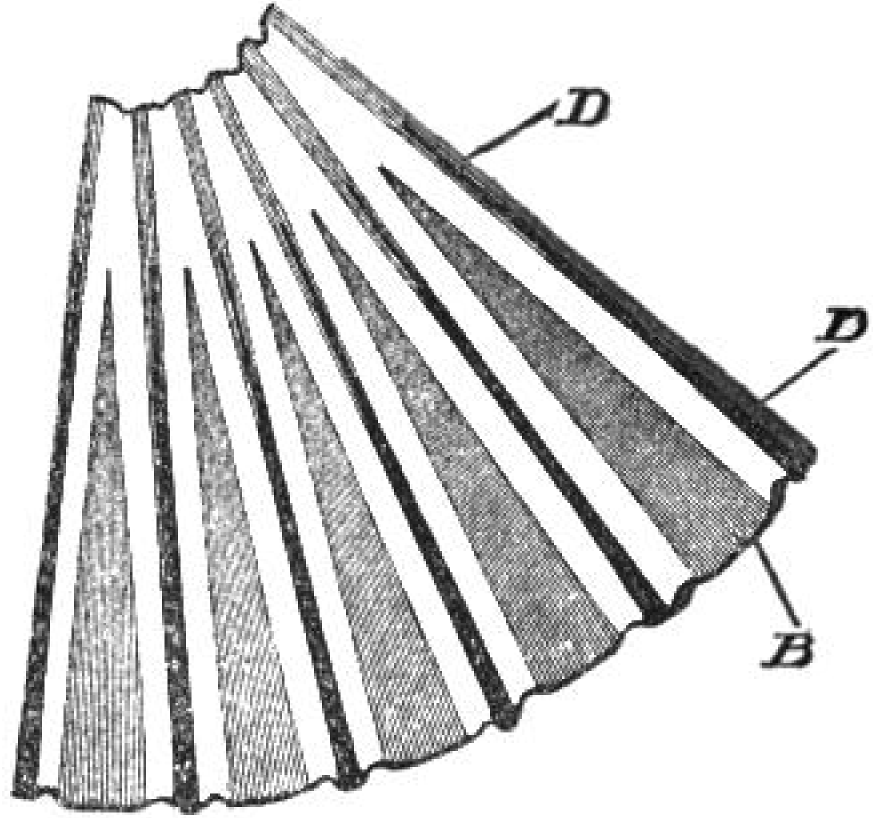



FIG. 2 (*R*) represents a relaxed fibril with a pin, *A*, sticking into the dark stripe. During its contraction (*C*), as the fibril simply shortens and thickens without otherwise changing its shape, the needle *A* is still seen sticking in the dark stripe.

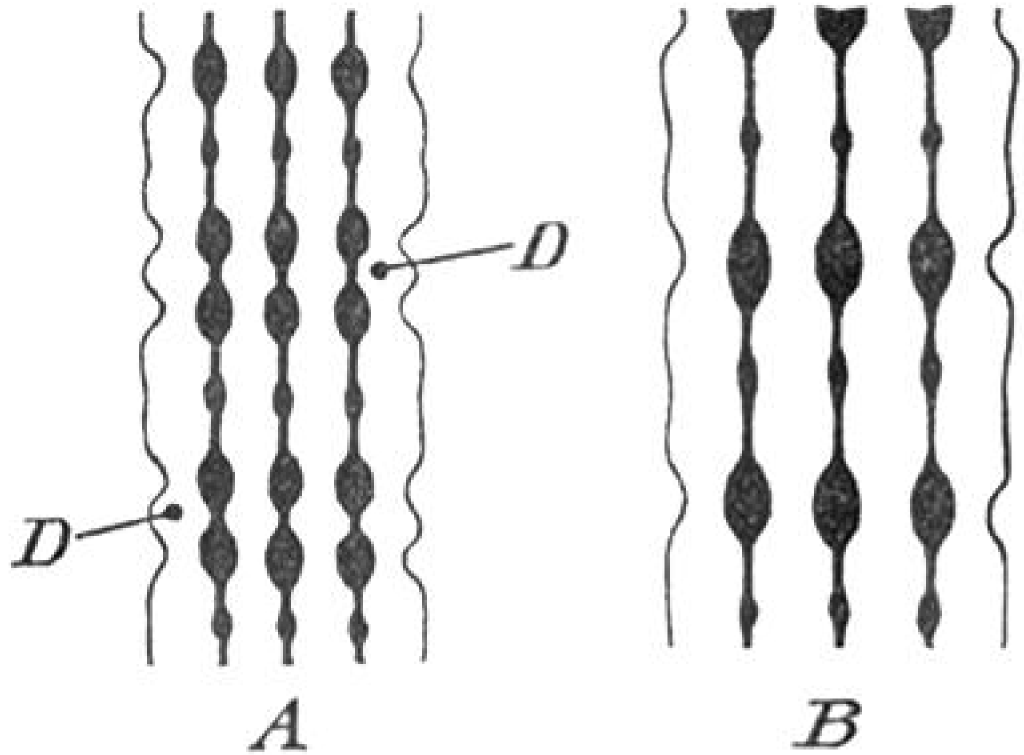
FIG. 3 (*R*) represents a relaxed fibril with a pin, *A*, sticking into the dark stripe, and another pin, *B*, sticking into the swelling in the position of Dobie's line. When contracting (*C*), the fibril is profoundly modified in shape, the pin *A* sticking into the clear stripe, and the pin *B* into the centre of the dark stripe.

FIG. 4.




A drawing of a living and contracted Crab's muscle, which has been bent round and artificially extended on its convex lower border. The transitions between the relaxed and contracted parts are well seen. Dobie's lines (*D*) gradually fade away as you pass to the contracted condition, becoming invested by and then replaced by the dark stripe of the contracted condition. The dark stripe of the relaxed part (*B*) fades away, and is replaced by the light stripe in the contracted part.

FIG. 5.

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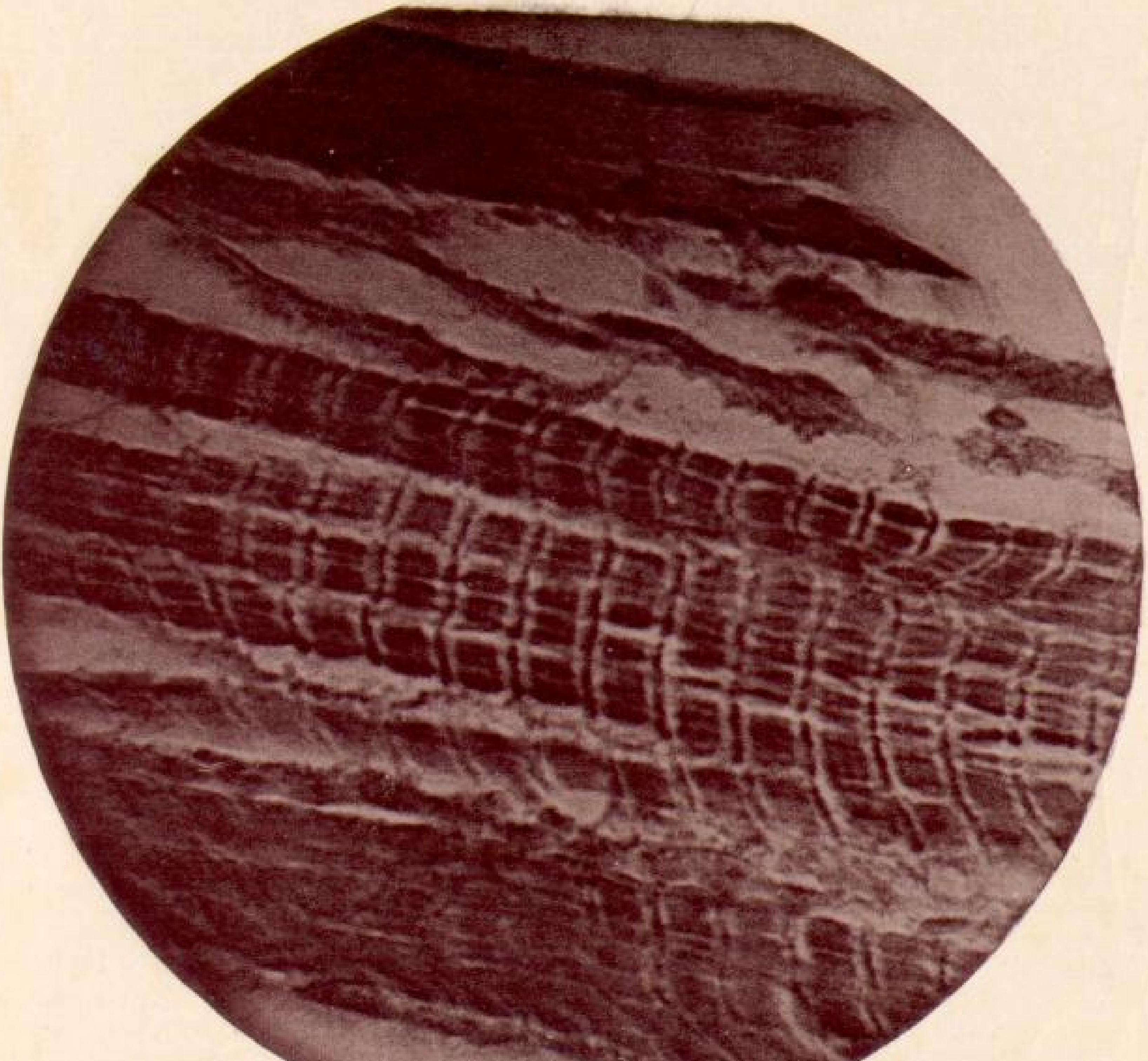

Fig. 1.

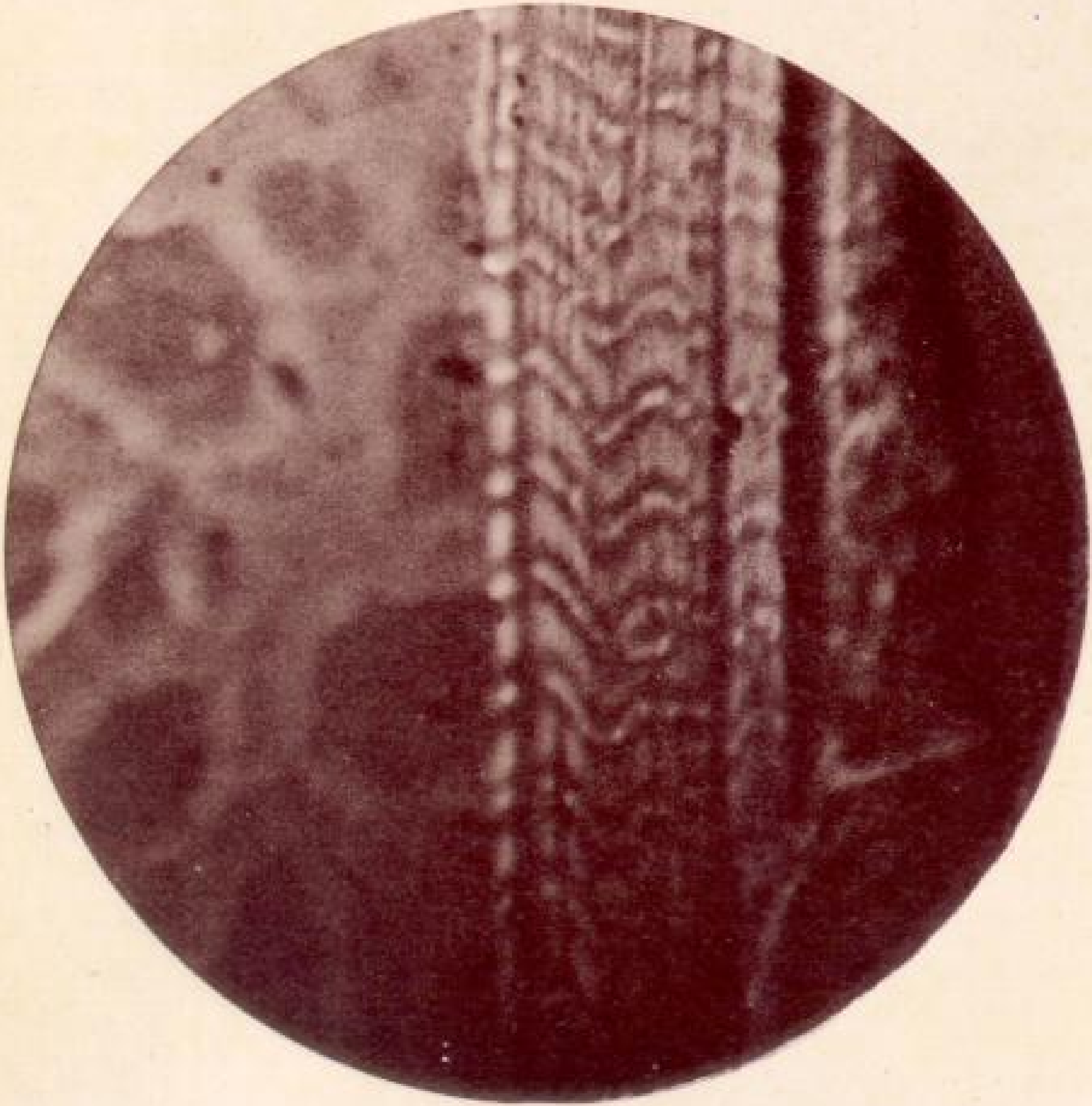

Fig. 2.

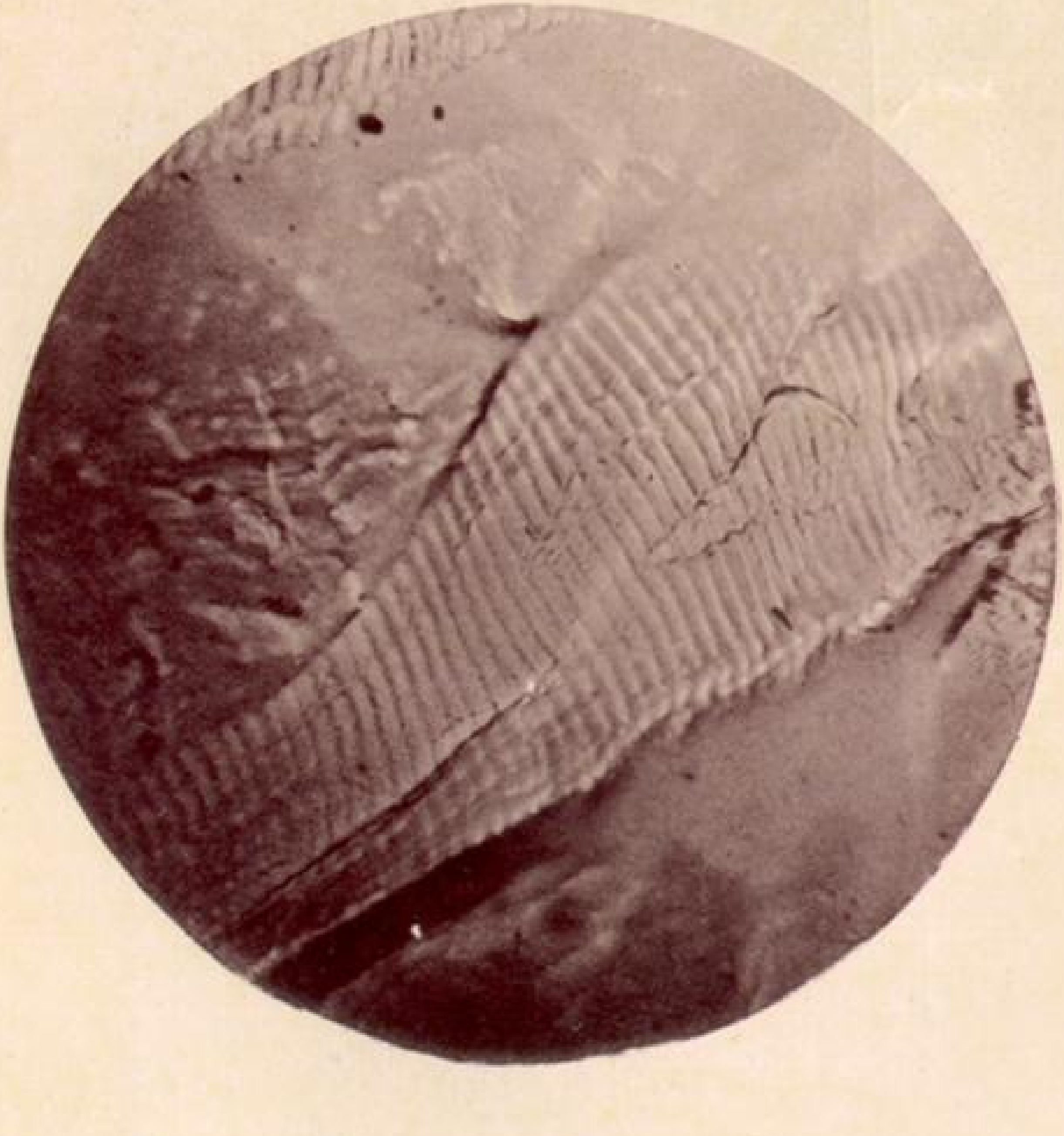

Fig. 3.