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IV.—*The President's Address.*

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(*Annual Meeting, 11th February, 1885.*)

PLATES IV.—VI.

THERE is a great temptation in the position which I have the honour to occupy to-night, to pass in review the more prominent and striking results of the work done in the year, touching the special branches of science to which the instrument, which we are constantly striving to bring nearer to perfection and to use perfectly, is ancillary.

The inducements to such a course have in fact rarely been stronger. The legitimate, patient, and scientific knowledge and use of the Microscope, has opened out not only unexpected areas of actual and potential knowledge, but has been the means of proving, at the same time, that this knowledge is most vital in its relations to the higher interests of our race and our civilization.

Nothing was more certain than that the work done in biology through the agency of the Microscope up to twenty years ago, was a distinct prophecy of far higher and more definite results.

Having touched the fringe of the minutest that was visible in living things, it was inevitable that a knowledge of all that was accessible concerning these, should be sought, and, if possible, found. The scientific certainty and value of the results would depend largely (1) on improvements of a thoroughly scientific character in the optical part of the instrument, and (2) on enlightened and patient manipulation of these.

We may, without undue exaltation, congratulate ourselves that by the combined ingenuity, knowledge, and skill of the Fellows of this and other Societies, and of the opticians, improvements of indefinitely great value have been effected in our lenses and

appliances, while the instrument itself has been advanced in perfectness.

But, on the other hand, the great doctrine of Charles Darwin, quickening, as it did, into new life and effort, every region of biology, gave immense impulse to work in all those departments of it which can only be dealt with and understood by means of our instrument.

The result has been the opening up of new knowledge, new methods, new directions and objects of research, and in one sense the formation of new biological sciences, within a couple of decades.

It would be impossible to follow this outspread of progress, due mainly to the Microscope, in the one science of biology; but, in only one direction, and within the years that lie closest to us, the value of this progress can be readily seen.

Our knowledge of the septic organisms has led an army of earnest workers to a relative and partial knowledge at least, of their pathogenic congeners; and in this, with our present resources, lie folded up as it were, indefinite possibilities for the future welfare of not man only—but man and beasts.

It is a little interesting to note in passing that an impetus was given to this question by the keen interest displayed some fourteen years, or more, ago, and onwards, in the question of abiogenesis *versus* biogenesis, and the masterly summary of the facts, and the inevitable deductions from them, made by Prof. Huxley in his address, as President, to the British Association in 1870.

Both biologists and, from a distinct point of view, physicists and chemists, as for instance Prof. Tyndall and Pasteur, continued to prosecute the inquiry; and during this series of researches two definite results were irresistible.

The first was that when the experiments were conducted with competent skill—as they certainly were in Tyndall's hands, in the laboratory of the Royal Institution, and on the Alps—it could be deduced absolutely that living things were never seen to arise in not living matter.

The second deduction, as made from the work effected by biological research pure and simple, was that the septic organisms, as such, do arise always, so far down as our researches have carried us, in the products, either genetic or fissipartitional, of like living things that had preceded them.

Abiogenesis was not a discoverable factor in nature's present condition, so far at least as our researches could carry us. And it is of moment to note that since Huxley's conclusions concerning Redi's doctrine of biogenesis were pronounced in 1870, not only has research in every direction of biological inquiry that could touch the question, confirmed that conclusion, but no facts with any

adverse bearing have been presented. Hence in the interval Huxley, taking into account all the factors of our ever-increasing knowledge of biological facts, affirms "that at the present moment there is not a shadow of trustworthy evidence that abiogenesis does take place, or has taken place within the period during which life has appeared on the globe";* and in another place, "the present state of our knowledge furnishes us with no link between the living and the not living."†

That there was a time when living matter did arise from matter not living there can be no question. How, is no present concern of science. With increased knowledge, deeper research, further penetration, even that question may be answered.

But the impossibility of present explanation is no scientific prejudice to the facts.

It is evidence of the deep insight of Darwin's mind that he nowhere suggested, or even hinted at, the operation of any unknown activity to give efficiency to the development of living things in all their incalculable variety. Out of the known and established phenomena of life he could deduce the evolution of all living forms. Granted the existence of living matter—primordial germs—he found no need of abiogenesis. And the long unbroken lines of life in definite forms, revealed by the geological and palæontological history of the earth, absolutely support this view.

The supposition that abiogenesis is a necessary sequence from the doctrine of evolution, has indeed no philosophical basis. The inevitable power of living forms, however lowly, to multiply without limit is enough. "In the eyes of a consistent evolutionist any further independent formation of protoplasm would be sheer waste."‡

But the investigations that in our more immediate times confirmed these conclusions, also gave us a greatly enlarged knowledge of that remarkable group of organisms known as septic. So much so, that the morphology of the monads and the bacteria may be said to be approximately within our reach, always remembering the range of our present optical aids.

But beyond this, the life-histories of many, of both the monads and the bacteria, are fairly known; and their physiological characteristics are being carefully studied. There is, however, a possible difficulty before us. The whole group of septic organisms must, as a group, form the subject of complete and exhaustive study. If this be not done, we are in peril of false conclusions on a practical phase of the subject, which touches the highest interests

* *Anat. Invert. Animals*, p. 39.

† *Biology*, *Ency. Brit.*, 9th ed., pp. 67-9.

‡ *Anat. Invert. Animals*, p. 38.

of the whole animal world, with man as the most profoundly concerned.

Of the septic series as such, I have a long-continued incidental familiarity. It is the monad group that I have of course, so far as I have been able to go, thoroughly studied. Yet it would be difficult for any one not a pathologist, and without the resources of a pathological laboratory, to claim to be in the modern sense a bacteriologist. But it may well be doubted whether that modern sense is a really useful and correct one. The bacteria are without much question a botanical group, and to be thoroughly understood must be studied as such. Whoever has studied the same field of septic bacteria for say a week or a fortnight without change of conditions, will know the strange complexity of relations that are seen to arise. I do not profess to interpret them—I have avoided the immense temptation to try to do so, the better to use such facility as I could command in the more complete working out of the monad group. But I feel assured that until this complexity of relation in common forms is understood, we cannot make really assured advance. How the bacteria are interrelated, how far they are mutable, and under what conditions; and whether functional changes are as readily, or more readily, induced, than morphological changes, are questions of the largest moment.

I should wantonly occupy your time if I attempted to call your attention to the fact that all this bears upon that special modification of the septic group only, known at present as bacteria or bacilli, which are pathogenic in their functions.

In some instances possessed of a specific functional power, they are the direct excitants, the positive virus, of definite forms of disease. To the study of these and their behaviour and life-histories there is a strong and a natural tendency. But it is to the specific and perhaps biologically highly modified form *alone* that all the labour is directed.

What is its relation to the whole group? Has it become modified from normal septic forms? if so, by what means; these are in the broad and deep interests of science even larger questions than the morphology and functions of a definitely discovered and demonstrated bacilli-virus.

Manifestly the work cannot be done by any one group of observers: the pathologists do wisely and well, to pursue their own department of the research with exemplary zeal: but the microscopical botanists have such a rich field of inquiry before them, and one so large in its importance, as has scarcely fallen to them before in the entire history of the Microscope.

That there is a direct and discoverable relation between the septic and the pathogenic bacteria it would be puerile in these days to doubt. What is that relation? In some instances, certainly, the

morphological difference between the ordinary septic form and the form that is pathogenic is by no means striking.

Yet the one is innocuous, the other deadly. The one is an organism with the power of growing enormously in simple chemical fluids containing nitrogenous compounds, such as tartrate of ammonia. The other refuses wholly to grow and multiply in this, but requires more complex compounds, as the proteids, for its nurture. Hence it is found in the blood or tissues of man and animals, and becomes a veritable *virus*.

The difference, however acquired, is functional immensely more than morphological. And the probable relation between the two forms is one of the most important by-problems of modern biology. It seems on the surface at least that we have here evidence of the operation of the great secular processes of the Darwinian law bringing about, in forms that retain a general morphological resemblance, a distinct physiological specificity.

I grant fully that no direct evidence, by experiment, of the change has been yet given us.

It is true that from the close general resemblance between *Bacillus subtilis* and *Bacillus anthracis* it was held that the only difference between them was physiological. This is in all probability not strictly accurate. But there is something yet to be learned in relation to the life-history of both forms, and certainly in relation to their mutability. Now Hans Büchner tells us* that he did by successive cultivations of the *Bacillus anthracis* under the influence of continued alterations of the nutrition, see it gradually change into the *Bacillus subtilis*; that in fact this deadly form by being passed through successive cultures at a temperature of from 35° to 37° Centigrade loses its pathogenic qualities, becomes, in short, bereft of its specific physiological character.

But more striking yet is the affirmation made by the same *savant*, that he has changed the harmless *Bacillus subtilis* into the deadly *B. anthracis* by similar processes.

To one who has fully comprehended the meaning and the operation of the Darwinian law, it will be at once apparent that there must be error somewhere in this matter.

If the law of actual variation, with all that is involved in survival of the fittest, could be so readily brought into complete operation, and yield so pronounced a result, where would be the stability of the organic world? Nothing would be at one stay. There could be no permanence in anything living.

This immediately impressed me nearly four years ago on reading this remarkable paper, and there can be no shadow of doubt that

* "Ueber die experimentelle Erzeugung des Milz-brand-contagiums aus den Heupilzen." SB. Math. Phys. Classen K. Bayer. Akademie d. Wiss., 1880, Heft iii.

Dr. Klein in his valuable papers on "Micro-organisms and Disease" has given a definite form and certainty to the utter improbability of Büchner's so-called results being in any scientific sense accurate.

But the fact that there is no evidence of any direct relation evolutionally between two such forms as *B. subtilis* and *B. anthracis*, the fact that there is no ready way either naturally or artificially of their being changed into each other, must not blind us to the fact, as biologists, that such an evolutionary relation in the past is eminently probable, nay almost certain. It may, in all probability must, have taken an indefinite time in the past to effect; but being once effected, the specificity is continued as in every other form by inheritance.

That *no* means now at work, or capable of being brought into play, is sufficient to-day to change the *B. subtilis* into *B. anthracis*, it is decidedly both in harmony with fact and philosophy to doubt; while it is of the utmost importance, if it be within the compass of human research, to discover what these conditions are.

Cognate with this question is the true interpretation of the real meaning and value of the attenuated intensity of the virus cultures of Pasteur and others. Is the diminished virulence of a given culture, which, as in the case of inoculation for anthrax, produces a mild form of the disease, leading to subsequent immunity, attributable to true biological change in the organisms brought about by changed environment, or is it a mere physical and accompanying chemical attenuation, consequent upon enfeebled nutrition, or extended dilution, of some element of the original virus outside the organism?

We can only answer, it is highly probable. What we know of the septic organisms leads us to infer that their tendency to adapt themselves to considerable variation of external conditions is evident; but this does not of necessity involve permanent change in the organism.

Now you are well aware that there is at this moment very much more that is similar to, or cognate with, what I have hitherto passed in rapid review, that relates to microscopic biology, and that stimulates, almost provokes, our consideration.

But in it all there is so much happily that is progressive, and *therefore* that is of necessity tentative, that I am fain to turn from these multitudinous and deeply interesting matters, feeling that I can better promote the interests of knowledge such as we seek, by the addition of definite facts, than by the most careful and extended review of the progression of knowledge, where much is unfinished, and some things are still in doubt.

Any President of this Society may be the more satisfied with such a course, who remembers the state of comprehensive efficiency

to which our Journal has been brought. The whole compass of such special or collateral knowledge as any of us may desire to reach, is either summarized or indicated for us there. Remembering this, I shall call your attention to a study I have been able to make of the life-history of a septic organism which I believe is hitherto new to science.

Some four years have been embraced in completing the work, and I have had the advantage of the very best and finest lenses which our recent advances in the theory and practice of the construction of high-power lenses have afforded.

I may remark, however, that for all work of the class I am about to detail, it is absolutely needful that, for continuous research, an air lens of high power should be used. You are well aware that both oil and water, as immersion media on the surface of the delicate cover, imperil, every moment, the fluid *beneath* the cover, and within which the subject of investigation is in constant motion.

For this continuous work I rely on a $1/16$, of great merit in all respects, as an air lens; on a $1/25$, which has very fine qualities; and on a $1/50$, which greatly surpasses, in all the qualities we seek, a lens of the same power by the same makers used up to four years ago.

The lens, however, on which I still rely chiefly for this work is a $1/35$, which for an air lens possesses qualities of the highest order.

But beyond the work of continuous watching, when the opportunity presents itself, there is the work of developmental morphology, of discovering all the details of the adult form, and of thoroughly demonstrating the changes that ensue in the completion of a life-cycle.

It is here that first, water immersion, and now, above all, homogeneous lenses have been to me of untold value.

Happily I have no need to deal with, even in review, the mathematical laws of aplanatic combinations, nor the diffraction theory of microscopic vision, and all that attends it, on which this vast improvement depends. In that I have been anticipated by competent masters.

But I ought to yield to none in the sense of obligation under which I rest for such splendid optical advance. With these new lenses I have gone over again all the critical parts of the work which I have had the honour from time to time to present to this Society, and others; and although I have discovered no error of interpretation, I have been compelled to see that many of the delicate points of detail, discoverable only by the most persistent effort and careful manipulation then, could be demonstrated with comparative readiness now.

The benefit arising from their use in the study I am about to

detail has been unusually great, especially since the organism is of extreme minuteness, and complex in the arrangement of parts.

I have been able to use in this investigation a $1/10$ homogeneous of large aperture, a $1/12$ of 1.47 N.A., a $1/25$ of 1.38 N.A., a $1/50$ of 1.38 N.A., and lastly for a little of some part of the work a $1/6$ of 1.50 N.A.

All these lenses were made for me by Powell and Lealand.

Some four years since I was examining cursorily an exhausted maceration of cod-fish which had decomposed in a broth extracted from the boiling of rabbits. The maceration had been for many weeks under examination with another object, but it had now become relatively devoid of the organisms it had contained, and there was, in the glass vessel in which it was contained, a considerable precipitate at the bottom. It had been kept during the long time it was under examination at a temperature of from 90° to 95° Fahr., and it was at the former temperature at the time of which I speak.

There were still a vast number of *Bacterium termo*, *B. lineola*, and *Spirillum volutans* in the fluid. With a purpose in no way connected with the issue, I examined with a $1/10$ homogeneous lens the slightly viscid and granular sediment; and in two or three of the many globules which I took out with a fine pipette, I detected the presence of an intensely active organism which appeared to present features and movement that I could not reconcile with any septic form familiar to me. It was so small that at first it appeared almost bacterial in its relations; but it was so involved in the slightly viscid mass, and withal its movements were so quick and constant, that it seemed almost impossible to fairly understand it.

To add to the difficulty it was manifestly very sparsely diffused in the fluid—or rather in the slightly viscid sediment—for it was wholly confined to this; and it was on an average only once in a dozen drops that two or three of the forms appeared.

I was examining it upon a cold stage, and as there was at least 30° difference in temperature between the stage and the fluid, such organisms as I was able to secure very speedily became enfeebled, and ultimately still. In the slower movements, and in the state of inactivity, I got a glimpse of characters that were, so far as I know, quite unnoted amongst the septic series.

By a simple arrangement, I adapted a hollow heating plate in the place of the principal part of the glass plate of which my continuous stage was made, as used with the temperature of the surrounding air, and I placed this heating plate in connection with a vessel of water kept by a mercurial gas regulator, worked with an unchanging gas pressure, at a temperature of 93° F.

The apparatus, as far as it is needed to reveal its purpose, is before you.*

My earliest and continuous difficulty was to find specimens for study.

As in former instances, I determined first to follow out the morphological details separately, and if possible exhaustively; and to fix on a given organism with all the details of its form known, from one stage to another, until I had been able to trace it from its earliest to its latest condition.

The first step was a careful measurement of its visible body sarcode. The body was of sub-oval shape, with a lenticular form on the upper and under surfaces.

The general appearance of the body alone is seen in figs. 1, 2, 3, plate IV., and on the measurement of twenty-five forms I obtained an average length of the 1/10,000 of an inch, and a breadth of 1/19,500 of an inch.

But when closely and carefully examined with a 1/25 oil lens, it presented the remarkable phenomenon of an organism so minute, with no less than six flagella, each one of which is nearly three times as long as the long diameter of its body.

It was soon evident that its movements were extremely varied, and some of them entirely unlike anything I had hitherto seen.

A not uncommon mode of locomotion was that of simple swimming, as seen in fig. 1, plate V., where the anterior and posterior flagella were in vigorous action, and the remaining four either trailed or participated but slightly in the direction of motion.

But a movement quite as often resorted to, was much more beautiful. The organism came to momentary rest much in the attitude seen in fig. 4, plate V., and fig. 10, plate IV., when by a sudden movement of the whole group of flagella, a series of wave-like leaps, reminding one of the movements of a shoal of porpoises, ensued, and was continued for from ten to fifteen undulations.

To actually make out the flagella and their movements required considerable "penetration" as well as the most delicate definition. A fine homogeneous 1/8 of Zeiss's greatly helped me in this matter; but at the last I reaped my most perfect results with the new 1/6 of 1.50 N.A. Indeed, I had the glass made with this special object; and the need of it will be seen more fully when I describe the essentially unique and characteristic movement of this monad.

In previous forms which I had studied, both with Dr. Drysdale and alone, I had observed and described forms that anchor the body by means of one or more long trailing flagella, which flagella are

* This will be illustrated in a subsequent part of the Journal.

made as it were the radii of a quadrant of a circle of which their fixed point is the centre; and by the rapid movement of the body to and fro in the circumference of the arc, the body was as many as thirty times per minute made to act hammer-like upon minute particles of decomposing matter. But in this organism, what was apparently the same result, was brought about in a manner wholly different.

The organism was never attached—had no power that I could discover of attaching itself for a moment; but by a free darting, down upon, and away from, minute particles of decomposing matter by the organism in large numbers, such matter was visibly, in the course of half an hour, reduced in size and altered in shape.

Fig. 9, plate IV., will give an idea of this mode of movement: *a* represents a minute particle of decomposing débris, and the arrows placed beside the organisms indicate the direction of motion.

Each one in turn, in a space of time occupying about two seconds, comes into contact with the particle *a*, and at once recedes to a distance of four to five times the length of the flagella, and instantly again darts upon the object, and this may be continued by given forms for hours.

No sight accessible to the human eye can be more fascinating or more beautiful than this, with delicate illumination and such definition as we can now obtain.

Fifty or even a hundred may be observed with ease, in one field, pursuing their untiring work. It is the more entrancing that it is apparently rhythmic, not like the measured march of a regiment, but the rhythmic movement of a peal of bells.

But the method in which the motion, with its manifest force, is brought about, is a matter of greatest interest, and the discovery of which is fraught with much difficulty.

The extreme minuteness of the organism, the large number of its delicate flagella, and the intense rapidity of its movement, are the three factors of difficulty.

It is absolutely needful first to familiarize oneself with its general movements before interpretation of this special action can be hoped for.

In swimming, and in the wave motion I have described, figs. 1, 2, 3, plate IV., and 1 and 4, plate V., all the flagella may be, with care, clearly seen; but in the act of darting down upon, and away from a piece of débris it is otherwise. Fig. 9 *b*, plate IV., shows one of the forms at the very moment of impact: *all* the flagella are behind, and (in this instance with the 1/12 objective, 1.47 N.A.) a spiral form is indicated in the arrangement of these flagella.

At *c* in the same figure, one of the forms, having accomplished impact, is receding. Two of the flagella now fall to the front.

But with much difficulty, though complete certainty, I was able to demonstrate a distinct movement of rotation.

The organisms start when at the greatest distance from the débris with the anterior and posterior flagella at an angle with the axis of the body, as in figs. 5, 6, 7, plate IV., and may, by close observation, aided with minute particles of carmine in the fluid, be seen to swirl rapidly from left to right, causing a screw-action, the middle four flagella presenting the spiral appearance seen in the figures; and the anterior and posterior rapidly approaching their final condition seen in *b*, fig. 9, plate IV.

In receding, the anterior and posterior flagella spread out as before, and swirl in the reverse direction, rapidly falling forward, as in *c*, fig. 9.

In this way the constant battering ensues.

Each of the drawings was made instantly, and they were made with different lenses. The whole of fig. 9 was made with the oil 1/25, 1.37 N.A.; so was fig. 11, plate V. Fig. 11, plate IV., was drawn with a fine water 1/16; fig. 6, plate IV., with the oil 1/50, 1.37 N.A.; while figs. 7 and 5, plate IV., were drawn with the 1/6 oil, 1.50 N.A.

A diagrammatic view of what I take to be the method of effecting this powerful movement, for accomplishing the final breaking-up of the last fragments of a decomposing mass, is seen in figs. 12 and 13, plate V.

The greatest perplexity at this point of my work was the relatively few forms I could find, and the difficulty of knowing in what way to promote their increase. I have not yet discovered this latter desideratum; but at a subsequent period I found the organism more freely, when the main difficulties of its life-history were overcome; and so I was able to complete the details here given.

But the incitements to proceed with the life-history of the organism, rather than linger over the details of its morphology and movements, were strong, for the drama partly observed was interesting in no ordinary sense.

From the large number of its flagella, I had hardly been prepared for fission as a feature of its life-cycle, although so universal in the group. But on steadily following one, and subsequently, in order to complete the observation, many others, I selected the free-swimming state, seen in figs. 4, 2, and 1, plate V., and fig. 10, plate IV. Soon, in the course of three or four minutes, I observed a sudden indenture, almost a constriction, in the middle of the body, as seen in fig. 3, plate V.; and directly this occurred, the flagella, previously acting together for the propulsion of the whole body, separated, and three of them acted in one direction and three in the opposite.

This pulled the constricted part out into a hyaline neck, as

in fig. 5, plate V. In the middle of this, at this time, there was a delicate line visible, parallel to its length. This became stronger as the neck lengthened, as in fig. 6. Still the dividing organism pulled its dividing parts away from each other, by the vigorous antagonism of the flagella; and the neck rapidly elongated, and split with considerable separation of the two lines of sarcode, as seen in fig. 7. But simultaneously with this there is a horizontal splitting of the cord *a*, fig. 7, which widely separates, and when the organism is seen on its dorsal aspect, shortly after this condition, it reaches that shown in fig. 8, where three flagella, of double length, are tightly stretched between the dividing organism, the upper one on a different plane to the two lower ones, and by a vigorous movement this is soon snapped in the middle, and two organisms quite complete are severed from each other, as in figs. 9 and 10, plate V., and fig. 12, plate IV.

It was impossible of course to follow both, but the one followed, in every case swam, after fission, with great vigour for from two to four minutes, when the first symptom of fission commenced again, and on the average occupied from four to five minutes in completion, from the moment when the first dorsal depression showed itself to the end of the process.

At this point the sparseness of my material greatly hampered me. I was deeply desirous of working out the history of the form to the end, while the actual material was before me. It was visibly diminishing; and although I had taken much pains to produce a new crop in other vessels, by sowing this one and subjecting it to the same conditions, I could have no certainty that it would succeed. Fortunately I did succeed twice afterwards in getting moderate quantities only; these were obtained respectively eighteen months and a little over three years subsequently.

It was not until about a year since that I was able to steadily follow the continuous fissions of three single and separate forms successively, by keeping one division continuously in view to the end. In one case there were forty-five and in two cases forty-two fissions, and the last divided form in each case entered upon the springing condition to which I have called attention.

It is in the next stage of the development of this organism that the real difficulty and the only incompleteness presented itself.

In all the monads hitherto studied, it will be remembered that besides the fissional stage, there is a state of *fusion* on the part of two individuals resulting in the production of spores: conditions no doubt representing in some sense the non-fertilized and the fertilized products of the micro-fungi.

But in all the cases known, the occurrence of this fusion is relatively rare. In this instance I had almost suspected that there

might be none, or that the small quantity I could obtain of the organisms would make the discovery hopeless.

But towards the close of the first set of observations my conviction that such a stage belonged to the organism was fully established, inasmuch as I met with the organism in the curious, but to me, extremely suggestive condition seen in fig. 14, plate V., where we have a distinct blending in part of two normal forms. The bodies were inverted in their relative positions; but in the upper one the flagella were, in a manner quite common to the group, fusing with each other, and with the body as at *a*, *b*, *c*, while the lower organism, *d*, was swimming with full vigour.

This observation was made, when nearly all the organisms had died out of my fluid, and although I observed this with the utmost care, I could only see for about an hour the slow absorption of the upper form into the under one, which increased slightly in size. But it fell to the floor of the stage, and after a couple of days observation it was abandoned as dead.

I had now to wait much over twelve months before I, in any of the very many cultures of the form which I had made, succeeded in obtaining it again. It is wholly confined to the nearly exhausted condition, of very powerful putrefaction, kept at temperatures ranging from 85° to 95° Fahr. and it is then extremely capricious.

My second find was however fairly copious at first; and I endeavoured to pursue the inquiry as to this method of fusion. Repeatedly I met with forms more or less approximately in the condition shown in fig. 14, plate V., and in every case save two (out of eleven) with the definite result I will describe immediately, but I was only successful three times in finding an earlier condition of this act of blending in all the period of my observation.

This earlier stage is fairly represented by one of the three, shown at fig. 15, plate V., where the two forms are in direct contact, and all the flagella are intact. Their movement was vigorous, and somewhat irregularly alternate; the flagella of one only being in action at once, until in one of the cases the flagella entangled and anastomosed with each other, and fused with the body as seen in fig. 14; then the other one assumed the horizontal position and swam freely; and slowly, the upper one was lost in the increasing sarcode of the lower until "each was melted into other." But there was no loss of swimming power. The motion was much slower, almost at times only a spinning on a perpendicular axis; but the body became vacuolated, and internally segmented in a rapid manner, but not easily discerned in the process from the movement of the mass.

In all preceding cases the sac of which this was the equivalent had been quite inactive at this stage: but in this case motion was unbroken, and in the course of from four to five hours a distinctly

granular mass was presented, as seen in fig. 16. The granules were more or less distinct, and were semi-opaque; but the flagella still were in active motion, and the whole mass was swirling and swaying with irregular movement over the whole field.

In earlier observations of this phenomenon, it was quite visible that when it had reached this stage, the mass grew gradually less; but how was not so readily discoverable, because it was so slowly effected.

But by careful arrangement it was subsequently seen with comparative ease that it was, as it swam, dropping a stream of semi-opaque granules, by no means difficult to see, as at *a*, *b*, fig. 16, plate V.

To study any changes that might happen in these was not a matter of difficulty. I did it chiefly with the dry 1/35; but where any points of special detail were to be made out I used the oil lenses.

The granules or sporules, or whatever we may call them, are at first semi-opaque: but in the course of fifteen minutes they lose this, and become transparent and highly refractive. Within a few minutes from the assumption of this condition they are visibly elongated, as at fig. 17, plate V. This elongation continues with general enlargement as at fig. 18. In about an hour it has reached the state shown at fig. 19, but it is very difficult to determine the earliest appearance of the setæ-like origins of the flagella which begin to show themselves between the stages shown in figs. 19 and 20, but after this the progress is very rapid; for in three hours more the organism passes through the stages shown in figs. 19, 20, and 21, and in the last condition begins its earliest movements; and rapidly appropriating food as it moves, it reaches the final condition shown in figs. 22 and 23, and was in several instances followed into the process of fission, depicted in fig. 24, and which constitutes a completing and re-entering of the life-cycle.

To complete our knowledge concerning this form so far as could be, I made, as in former cases, a series of experiments on the thermal death-point of the germs.

The apparatus I employed for this purpose is that which I have before employed for the same purpose, and which I have described, both to this and the Royal Society.* It enables an examination of the immediately emitted germ to be subjected to such thermal conditions, in fluid, as may be desired, and the germ to be subsequently studied under any power, without the slightest disturbance, and in normal conditions.

From the nature of the spore-sac, and the slow mode of

* See this Journal, iii. (1880) pp. 1-16, and Proc. Royal Society, xxvii. (1878) p. 332 *et seq.*

emission, I was enabled to use this apparatus in its simplest condition, and so to ensure less complex results.

The death-point of the adult was first tested. I found that it was not killed under a temperature of 146° Fahr. This was from 4° to 6° higher than any other form I have studied.

After ten separate tests, carefully worked out, I found that the highest point which the spores or germs could endure, and still germinate, was 190° Fahr. After this point was passed the form never appeared in the closed cell. That is to say, that the spores seen to be deposited in the specially prepared cell, did germinate after being raised to any temperature intermediate between 146° and 190° Fahr., but beyond the temperature of 190° the spore lost its power of germination.

This is the eighth of these most interesting forms that, either in conjunction with my colleague or alone, I have been able to carefully follow through their principal developmental changes.

It may be well to indicate briefly some of the broader aspects of the study as a whole.

And first, whether these organisms are animal or vegetable, is without doubt an interesting question. But the more completely we know the organisms the more difficult does the determination of this question become. It may be taken as absolutely certain that, in common with the septic bacteria, they can live, thrive, and multiply indefinitely in fluids totally devoid of proteinaceous matter, and composed wholly of mineral salts and tartrate of ammonia—as Cohn's nutritive fluid. On the other hand, they are probably even more prolific in putrefactive animal and vegetable matters.

But no definite inference can be made from this; for a form so essentially animal as *Paramæcium aurelia*, and still more abundantly *Stylonychia pustulata*, will live and rapidly multiply with them in the same simple nitrogenous compounds.

To argue the vegetal character of these forms from their plant-like ability to produce protein when supplied only with carbonic acid, ammoniacal salts, some mineral salts and water, would be, with the fact I have stated, to reduce the genus *Paramæcium* to vegetal forms—a result which few thorough biologists could sanction. On the other hand their normal ability to dwell in putrescent animal and vegetable fluids, places them on a par with *Paramæcium aurelia* on its animal side; and thus their double capacity in this matter appears to give them an intermediate position, touching almost equally the animal and the vegetable series.

Not less interesting in its general biological bearing is the whole process of fission in this group, especially when its varieties are taken into account.

In some instances, as in the case we have considered to-night, the division of the sarcode is simple; it is, so far as our power to see can carry us, a mere cutting of the sarcode into two independent parts. But to secure the independence of each, the most complex arrangements for the supply of flagella have to be provided. The ease and simplicity with which this is wrought is indeed striking.

On the other hand, there are forms in which the division itself is a mere equi-partition of the sarcode of the body, devoid even of a discoverable nucleus, and the pulling out, at the point of division of the organism, of a simple neck of protoplasm into a single fibre making new flagella for the two separated parts.

This is shown in fig. 1, plate VI., *a* being the normal form, *b* the earliest discoverable state tending to fission; the constriction deepens, and each half by individual activity recedes from the other, as in *c*, *d*, pulling out the fibre *e*, which snaps in the middle and each half goes free.

But not only have we instances of simplicity and of complexity in the fissional provision of flagella; but there may be increased complexity in the process by the clean division into two of the nucleus.

In the form shown in fig. 2, this will be readily seen. *a* is the adult form of the organism. It can swim freely; but is usually "anchored" by a "trailing flagellum" *b*. It has a large hyaline nucleus *x*.

The first movement towards fission is a partial division of the front flagellum, as at *e*; then the sarcode of the body begins to divide as at *d*, accompanied by a splitting of the upper part of the trailing flagellum *d*. The incision in the sarcode deepens; and with this the vivid nucleus *x* shows an incision in its upper part. Rapidly the two halves of the dividing body are as it were torn from each other, the front flagellum is wholly divided, *f*, *j*, *k*, the trailing flagellum nearly so, as at *m*, *n*, *o*, *p*, and the nucleus is quite divided: immediately after which the two perfect halves are free.

This is a complex but exquisitely beautiful fission.

But there are two of the forms that present almost the same problem of how to accomplish a multiple fission; and each solves it in an independent manner. Their mode of multiple division is seen in figs. 3 and 4. In fig. 3, *a* is the simple normal form. It swims for a couple of minutes, then assumes a rounded discoid appearance, and becomes still, as at *b*, the flagellum *c* melting into the sarcode.

A white cross suddenly appears as at *d*, which is rapidly followed by another at right angles to the first; then it twists into

a turbinated shape *e*, which is at once followed by quick and continuous internal movement, which results in the breaking up of the mass into a condition shown at *f*, where there is no trace of investing membrane, and the parts are in a state of movement upon each other for some twenty minutes, when the whole breaks up into perfect, but minute adult-shaped forms, with complete power of motion, as at *g*.

In fig. 4, *a* is the adult form. It is always invested with a hyaline coat; after swimming for awhile the interior sarcodē constricts as at *c* in *b*; the constriction deepens, as at *e* in *d*; and the two halves separate within the membrane, as two oval bodies, *g*, *h* in *f*. Subdivision goes on rapidly within, until sixteen perfect oval bodies with minute flagella lie within the mother body, and at length break the investment and go free.

Here we have the same result accomplished, with and without an investing envelope: and in both cases after a long-continued increase by each individual thus separated by fission, there is a perpetuation of the form by a fertilizing process and spore.

There are two other forms that present remarkable phenomena in self-division. They are shown in figs. 5 and 6.

In *a*, fig. 5, we see the common condition of the largest of the septic organisms I know. In the course of a few minutes, if followed, it is seen to lose its sharpness at the end opposite the flagella, and is amœboid, in the course of a couple of minutes presenting an appearance shown at *b*. The amœboid condition is seen at *d*, but in this state the root or base, out of which the four flagella spring, divides, and two pairs of flagella, each independent of the other, recede from each other, and the nucleus partially splits, as at *c*. The amœboid condition in a short time ceases, as at *e*, the flagella rapidly recede from each other, and the nucleus almost wholly divides, seen at *e*, *f*; at this point there is a sudden flattening of the organism from before backwards, and a lateral lengthening, as at *g*; the flagella still recede, as at *h*, *i*, and a constriction appears, as at *k*. Soon the flagella are at opposite poles, and pull against each other; the neck of sarcodē is drawn out, as at *l*, and with it each half of the nucleus into its separate part. A moment or two after it has reached the state shown at *l*, a vigorous movement snaps the union, and two perfect forms are free.

The other and final instance is seen at *a*, fig. 6. The form has an anterior and two lateral flagella, and swims with supreme grace.

The first indication of fission is a delicate slit in the anterior knob of the body *b*, and with the opening of this the front flagellum splits.

At the same moment a slit *d* appears at the posterior end, and with this an incision in the nucleus. Both slits rapidly open; two anterior flagella are formed, as at *e*; the nucleus wholly divides, as at *f*, and soon only a neck of sarcode connects the dividing halves, as at *g*, *h*. Pulling in opposite directions then takes place, lengthening the cord, as at *i*, until it reaches the fineness, and double the length of the flagellum required for each divided form, as at *k*, when it snaps and each is free.

The mystery of all this simplicity of vital movement is deep; and although we can observe and in accurate manner record the process, its *modus operandi* is far beyond our present grasp.

One thing is certain, on this rapid power of self-multiplication depends the entire utility of these organisms, and in this function of self-division it would appear that they have reached the highest point of vital development. Lowly they are—we know of nothing living that is lowlier—but in the processes of vital evolution, amongst the lowly and simple as amongst the highly organized and most complex, we find the perfection of concurrent adaptation.

One other feature in these minute organisms claims a note. They are subject to no caprice; after twelve years of close observation I am convinced that the vital processes are as orderly, rigid and immutable as in the most complex organisms. Their life-cycles are as clearly definable as those of a crustacean or a bird. No vital phenomenon not to be found amongst higher and larger organisms, is discoverable here. Only the methods of specific mutation resulting from the secular processes enunciated in the Darwinian law are in operation.

But the action of these processes is certain; and from the rapidity of their increase, the shortness of their life-histories, and the ease with which vast multitudes may be studied, these organisms afford a splendid field—by changes of environment—for proving the action of the Darwinian law. But this must be a work in which it would be vain to enter, unless, relatively speaking, vast time were given. For, immense as their multitudes are, and rapidly as generation succeeds generation, and easily as changes of relative minuteness may be seen, it is manifest to a close observer that definite and discoverable change, in response to a marked change of environment, may be brought about; and there can be no doubt that the tendency to vary is fully operative in the group; and with it a tendency to conserve useful variation. This to an interesting extent I have proved, and shall hope on a future occasion to give you the evidence afforded.

Meantime, however, I may remark, that so far as my observation has extended it is never in the process of fission that variation

occurs—the divided organisms are always normal. It is, at least so far as my observation has extended, only in the forms arising from spore that there is any tendency to vary.

EXPLANATION OF PLATES.

PLATE IV.

Figs. 1-4 show the general form of the organism with its six flagella, and the attitudes assumed in ordinary movement.

Figs. 5-9 illustrate the special movement of the organism when engaged in large numbers in battering minute quantities of decomposing matter, as *a*, fig. 9. The arrows indicate the direction of movement with the condition of the flagella. Figs. 5-8 and 11 are illustrations of the appearance of the flagella when the various stages of the battering are watched.

Fig. 10.—Attitude of the organism before fission.

Fig. 12.—The final stage of fission before actual breaking into two.

PLATE V.

Fig. 1.—Appearance of the form when in ordinary motion.

Figs. 2-8.—Various stages of the act of fission.

Figs. 9, 10.—Two forms separated by fission—the result of the division of the organism in the condition seen in fig. 8.

Fig. 11.—A drawing of the organism as it appears when seen in the best conditions for observation in the act of "battering."

Figs. 12, 13.—Diagrammatic view of what is in all probability the arrangement of the flagella in the state of motion shown in fig. 9, plate IV.

Figs. 14-24.—Various stages in the genetic fusion of two forms issuing in spores (fig. 16) and the development of young forms (figs. 17-24).

PLATE VI.

Various modes of fission in different forms of the monads already worked out.