

## CHAPTER X.

SPONTANEOUS GENERATION (*concluded*).

Facts with regard to unboiled fluids and tissues: Mr. Lister's facts with regard to milk: my own experiments with animal tissues. Present state of the question—Dr. Bastian's latest standpoint: Liebig's doctrine. Can organic molecules derived otherwise than from pre-existing bacteria give rise to bacteria? Firstly, there is nothing unreasonable in looking on these particles as bacteria or their germs: seeds carried by air: excessive minuteness of the germs of bacteria: Mr. Lister's experience with micrococci. Secondly, there is no direct evidence that organic molecules not derived from bacteria can give rise to bacteria: facts with regard to unboiled fluids and tissues—conclusions from these facts: Paul Bert's experiments: Cazeneuve and Livon: Pasteur: Dr. Bastian. Thirdly, there is no evidence that active organic molecules (chemical ferments, &c.) can resist heat more than living things can: effects of heat on ptyalin, pepsin, &c.: my own case of difficulty in sterilising milk: Wyman's fact as to an alga living at 208° F.

WITH the great mass of evidence existing against the doctrine of spontaneous generation, I should have no hesitation in casting it aside. But much stronger evidence remains to be alluded to; viz., the behaviour of unboiled fluids and tissues when preserved with certain precautions.

I have already referred to these experiments in a former part of this work, and I may now state that where I have said that fermentation was absent, I might equally well have said that organisms were absent. I have mentioned that blood, milk, and urine could be preserved, unboiled and unacted on by chemical reagents, for an indefinite period, without undergoing fermentation, and, I may now add, without the development of organisms.

Of these I may revert for one moment to Mr. Lister's experiments with milk. Mr. Lister, in his attempts to obtain pure unboiled milk, found that in none of his flasks did he obtain lactic fermentation, but that in all but two (out of

some 50 or 60 flasks) organisms of some form or other occurred, these organisms being in many flasks of totally distinct kinds.

Now these results prove a great deal against the view of spontaneous generation. Two flasks had no organisms at all, therefore milk does not spontaneously generate organisms, does not naturally contain them, has no inherent tendency to undergo fermentation. Then milk which has never been in the dairy does not undergo lactic fermentation; hence the cause of this fermentation is something coming from the dairy, not some form of organic molecule present in the milk. Then not only the fact that in two flasks no organisms developed, but also the great variety which appeared in the different flasks prove that they could not have developed *de novo*. For if we have twelve flasks under the same shade, each containing the same specimen of milk taken at the same time, under the same conditions, any change occurring in one, due to something inherent in milk, or due to some physical force or combination of physical forces acting on it, would have occurred equally in all. But when we see one specimen remaining without organisms, another having a pigment micrococcus, another having bacteria, another fungi, and so on, we must conclude that the appearance of these forms cannot be due to anything inherent in this milk, but that it is due to something which has entered the milk from without.

My own experiments on milk, referred to before, bear out Mr. Lister's statements in every particular, and bring additional evidence, in that they show that a much larger proportion of flasks may be preserved if attempts be made to render the particles in the air, or at least on the udder of the cow and the hands of the milkmaid, inert by the use of carbolic acid or other antiseptic.

Cazeneuve and Livon's experiments on urine have been already referred to, and will be further discussed under the head of fermentation, when they will be found to furnish much additional evidence against the view of spontaneous generation.

After referring to blood, milk, and urine, I also mentioned the experiments on grape juice by Van der Broeck, Pasteur, &c., and on vegetable tissues by Roberts, and I then passed on to

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my own experiments on animal tissues. I described the method I had employed, how the tissues were introduced into and preserved in flasks containing cucumber fluid, and how they remained unchanged, and, I may now add, without any development of organisms. That no organisms developed I further ascertained by testing the beakers. By transferring, by means of a pure syringe, a drop of their contents into a flask containing pure cucumber or meat infusion, I found that no organisms developed in the latter, proving that none were present in the former. That organisms would have been found by this method, if they had been present in the fluid, is shown by the fact that, if bacteria were injected into the animal immediately before death, they were found in the beakers and in the flasks. I have also stained the granular matter present in these beakers with methyl violet, according to Koch's method, and have failed to find any organisms (see Plate V. fig. 34).

From all these facts it may therefore be regarded as certain that organisms do not originate spontaneously, in the sense formerly held, under any circumstances, but that they appear in animal or vegetable fluids and tissues as the result of the entrance of *solid particles* into these after their exit from the living organism. That the question has narrowed itself to this, that it is now no longer a question of spontaneous origin of organisms in fluids which have been severely boiled, but that it is a question as to whether *some of the particles* which get into fluids and which resist heat may not be organic molecules not necessarily derived from pre-existing bacteria, or whether they are bacteria or their germs, is evident from what has gone before, and at last Dr. Bastian has formulated similar views in his paper in the 'Nineteenth Century' for February 1878. He says:—

'The modern opponents and supporters of the doctrine of spontaneous generation have always been principally concerned with two sets of problems, (1) As to the nature of the material in the air, the access of which is so apt to induce fermentation in suitable fluids; (2) As to whether some degree of heat below 212° F. can be proved to be always sufficient to destroy

the life of different kinds of living matter in the moist state, but especially that of bacteria and fungus germs.

'In regard to the first set of problems it has been generally agreed for some time that the air contains some germs of living organisms, but that what proportion these bear to the much more bulky, and probably more numerous, organic particles and fragments resulting from the breaking up of previous living matter of various kinds, is uncertain. It has also been generally admitted that any living organisms or germs which chanced to fall from the air into suitable fluids would initiate fermentation or putrefaction therein. The question really requiring to be solved has always been (though it has not been uniformly recognised) whether mere organic *débris* from the air, either in the form of particles or of larger fragments, could or could not also bring about such changes in suitable fluids.'

This view corresponds very closely with the position which Liebig was ultimately driven to take up on the subject of fermentation, and Dr. Bastian says: 'It is Liebig's doctrine which legitimately suggests the doubt above mentioned in regard to the possible potencies of atmospheric particles other than actual germs . . . . If it has been shown that the appearance and increase of the lowest living particles are always a correlative of these processes (fermentation and putrefaction), Liebig's view, if it be true at all, *must be true for the whole of the processes*<sup>1</sup> which are essentially included under the term fermentation.' It will thus be seen that Liebig's theory and Bastian's recent views stand or fall together, and that the facts against one tell equally against the other. Just now I shall only deal with Bastian's views, but their intimate relation with Liebig's theory must be borne in mind, so that the full significance of the facts opposing the latter may be recognised in their bearings also on the former.

Dr. Bastian later on says: 'I go no further than to say that in the present state of the evidence bearing upon the subject I regard the hypothesis of spontaneous generation as the most logical and consistent interpretation of the facts which are at present known.' We must therefore consider what the evidence on this subject is, and whether there are any grounds for accept-

<sup>1</sup> The italics are mine.

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ing the theory that the organic molecules which give rise to bacteria originate from matter other than pre-existing bacteria, rather than the view that they are directly derived from pre-existing bacteria, in other words, that these particles are nothing more or less than bacteria or their germs.

I. Firstly, *there is nothing unreasonable in looking on these particles as bacteria or their spores*; indeed, Dr. Bastian admits that some of them are derived from bacteria. But if *some*, why not *all*? Because some resist heat more than others? We shall return to this presently.

We know that the seeds of plants are often wafted by the air, and this is more likely to be the case the lighter the seed, and therefore it is generally admitted that the seeds of fungi, large or small, are carried by the air. Indeed, both Pouchet and Pasteur found and recognised the spores of fungi in air dust.<sup>1</sup>

Further, bacteria propagate by fission in fluids, that is to say, one bacterium develops from another. This process has been often observed. Then, again, some forms of bacteria have been found to produce spores, notably forms of bacilli. Thus the spores of *Bacillus anthracis* have been fully described by Koch, and still more recently the occurrence of small sporules of excessive minuteness has been described by Ewart.<sup>2</sup> Ewart has also asserted that other forms of bacteria, especially of *Spirillum*, produce spores in like manner. If then in fluids bacteria always grow from bacteria, if some bacteria like fungi produce spores, why refuse to believe that the particles in the air which give rise to bacteria are these organisms themselves or their spores, just as the particles which produce the larger fungi have been shown to be their spores? The excessive minuteness of the bacillus spores renders them difficult of discernment in fluids under the microscope. When dried and shrunk we

<sup>1</sup> In December 1880, having collected a quantity of laboratory dust by aspiration of the air through gun cotton for several weeks, I dissolved the gun cotton in ether and alcohol, and collected the dust on a glass slide: this dust was stained with methyl violet according to Koch's method (see Chapter XII.), and a drawing of it is given in Fig. 35, Plate V. It will be seen that one or two bodies are present which are indistinguishable from bacteria and micrococci, and these bodies have been stained by the methyl violet.

<sup>2</sup> See *Proceedings of the Royal Society*, 1878.

should expect them to be hardly visible; and if the spores of such large organisms as *Bacillus anthracis* are hardly visible, how much less likely are we to find the spores of organisms which themselves can be seen only with difficulty? It seems to me that it would be a very extraordinary thing if in the case of these *minute organisms alone*, the particles which gave rise to them were not derived from pre-existing forms.

But when we find that under certain circumstances the same form of organism originates from these particles, as we should expect were the latter spores, the case is made still stronger. This latter fact will be more evident when we come to consider the relations of these bodies to fermentations, but I shall here mention one experience related by Mr. Lister.<sup>1</sup> In some experiments on the growth of a fungus in urine, a form of micrococcus, which he terms *Granuligera*, constantly appeared in the urine. He found that these were really organisms, from an opportunity which he had of watching their growth. He then says:—‘About this time, my study suffered from an epidemic of *Granuligera*. I could not now perform the same experiments with the same success as in the first instance: any that I tried was sure to be followed by the development of this pervading organism. I eluded it, however, by continuing the investigation in a room at the top of the house, which had been for a considerable time unoccupied. Here the results of the experiments corresponded with those originally obtained in the study.’ In this experiment there cannot be the slightest doubt that the particles which gained access to the cultivating fluids were these micrococci or their spores, if they have any. It could hardly have been some form of organic molecule not derived from these organisms which always gave rise to exactly the same forms.

Such are some of the facts which show that it is not unreasonable to look on these particles as bacteria or their germs; and Dr. Bastian, as I have already said, admits that some of the particles are derived from pre-existing bacteria.

II. Secondly, not only is it not unreasonable to regard these particles as bacteria or their germs, but *there is no direct evidence whatever that organic molecules not derived from bacteria can give rise to bacteria*.

<sup>1</sup> See *Transactions of the Royal Society of Edinburgh*, 1875.

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Look at all the facts I have brought forward as to unboiled fluids and tissues. I have shown that the most diverse organic fluids and tissues may be preserved for an indefinite period, without the development of organisms so long as the particles in the air and on surrounding objects are excluded. And yet these substances are full of organic molecules derived from previously living structures.

Cut a piece of liver, kidney, muscle, or other tissue from a healthy animal not yet dead, and place it, with precautions to exclude atmospheric dust, in a flask containing cucumber solution. Here you have at first in the fluid and in the tissue *living* cells and *living* organic molecules—and we know, as in the case of muscles, that life may be retained for some time—and yet no organisms appear. These living organic molecules of all kinds do not give rise to bacteria or any other form of life—they die.

Again, when the same specimen has stood for some time, we have a highly putrescible and fermentescible material containing *dead* organic molecules of all kinds; and yet no life appears, and such flasks may be kept for an indefinite time without the development in them of life.

But again, these organic molecules are not only dead, but *decaying*. They undergo slow oxidation—what Liebig has termed *eremacausis*. And yet they do not develop into or induce the appearance of living forms.

Organisms only develop in these fluids and tissues when bacteria or their germs or when atmospheric dust is introduced. Whence, then, are the hypothetical organic molecules derived? If they do not develop life in such fluids as cucumber or meat infusion, or in meat itself, under the conditions described, how is it that they come to do so after having floated through the air? Is it that they have acquired new properties in the air, or is it that the organic molecules (not derived from pre-existing bacteria) which, falling from the air into the fluids or on the tissues, give rise to living forms, are specially manufactured in the air? One or other of these conclusions must be accepted, and I say they are both equally untenable.

And when I come to speak of the relations of organisms to fermentations, more especially to fermentations occurring in wounds, I shall bring forward evidence of a similar character.

Thus I take flasks containing pure, highly putrescible fluids, such as meat or cucumber infusion, or milk, and I introduce into them pus, blood, or serum from wounds, and place them under the most favourable circumstances for the development of organisms, and yet no organisms develop. The same reasoning applies here as in the case where larger living masses, as liver, have been introduced into similar infusions. On the other hand, if bacteria or micrococci are present in the discharge (and I confirm this by microscopical observation), organisms develop in these flasks, and organisms of precisely the same morphological characters as were present in the original discharge.

But let us glance for one moment at Paul Bert's work.

He subjects such substances as saliva, pepsine, myrosin, emulsin, &c., to high pressures, say twenty atmospheres, and he finds that when the normal pressure is restored, these substances have not in any way suffered, as regards their fermenting power.

On the other hand, he places putrefying or fresh meat under a similar pressure, and after restoring the normal pressure, he finds that if he excludes fresh atmospheric particles, putrefaction is in the first case arrested, and in the second, never takes place. Organisms never again develop unless fresh dust is admitted.

From the first set of experiments we see that organic molecules when they have any power of acting are not deprived of it by high pressure, while from the second we learn that under similar circumstances life is destroyed, and no new life appears. And yet this cannot be because the power of generating organisms, which organic molecules are supposed by Dr. Bastian to possess, has been destroyed by the high pressure; for, as we see in the first experiment, dead organic molecules, as distinguished from living ones, retain their powers even under this pressure. The organic molecules, then, which originated the bacteria were living molecules, but *not merely any living molecules*, as is evident from the liver experiments, *but living molecules derived from pre-existing bacteria*.

Then, again, Cazeneuve and Livon's experiments, which will be detailed presently, prove exactly the same points. They show that it is not organic molecules in urine or in the wall of

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the urinary bladder which originate bacteria, but that it is organic molecules derived from the dust of the air; in other words, derived from pre-existing bacteria.<sup>1</sup>

All the proof which Dr. Bastian attempts to adduce on this point is to bring forward the opinions of several eminent metaphysicians as to the first origin of life on this globe. But with all respect for these great minds, I do not see one particle of *proof*, but merely metaphysical speculation, in the extracts quoted. Dr. Bastian removes the question to the first origin of life, and at once throws aside the creation view as untenable. I do not intend here to enter on this question, but I for one am not prepared to go so far.

III. Thirdly and lastly, we have *no evidence that active organic molecules can resist heat more than living things can.*

Let us take the so-called 'unformed' ferments, which consist of active organic molecules. The action of saliva on starch is favoured by a temperature of from 35° to 40° C., but its amylolytic activity is permanently destroyed by heat, even below the boiling point, applied for a few minutes.

The same is true of gastric juice; 'at temperatures much above 40° or 50° C. the action of the juice is impaired. By boiling for a few minutes the activity of the most powerful juice is irrevocably destroyed.' (Foster.) And so with pancreatic juice, &c.<sup>2</sup>

I have previously discussed at length the errors in experiment and observation which have led to the belief that organisms can develop in materials subjected to a *moist* heat for a sufficient length of time. And I have quoted Dr. Roberts's refutation of the experiments with alkaline fluids.

I stated on page 193 that I had only met with one instance, in an exceptionally long experience, of difficulty in sterilising an organic fluid. This fluid was milk got from a particular shop near the Strand. When I got fresh milk from

<sup>1</sup> Again, Pasteur by his experiments of opening vessels in different situations has distinctly shown that *all* particles of dust do not cause development of organisms; for undoubtedly organic *débris* did get into all his flasks, especially those opened outside, and yet it was only when certain special particles entered that organisms developed.

<sup>2</sup> Compare also Bastian's views on the *Disruptive Agency of Heat*, referred to at page 190.

a dairy in Albany Street, I had no difficulty in rendering it barren by immersion in boiling water for a few minutes. But the milk from the shop near the Strand has been immersed in boiling water even for three hours, and yet organisms developed in it. It is to be noted that in all cases the bacteria were identical in form and in the effect which they produced on the milk: they were a form of long bacillus. Now here we had some particle which got into the milk and caused a special change in it, and a constant development of a particular and easily recognisable form of organism—an organism which, moreover, has been found by other observers to resist heat, and especially dry heat, in an extraordinary manner. Is it reasonable to suppose that the particles which gave rise to these organisms were organic molecules derived, goodness knows from what, or specially manufactured in the air of this shop? Or is it not more reasonable to suppose that the air was infected with some form of bacterium or its spores, just as in Mr. Lister's case with *Granuligera*, and that this special form or its spores possessed the same resisting powers which it has been found to have in other parts of the world. For I may add that the spores of bacilli are stated by all who have investigated the subject to be possessed of extraordinary resisting power.

After all it is not a matter of great surprise that an organism should resist a heat of 210° F. (that has been ascertained by Mr. Lister to be the temperature of milk immersed in this way), for Prof. Jeffries Wyman tells us of a form of alga which lives normally in water the temperature of which is 208° F.<sup>1</sup>

With this great mass of evidence, and I could have multiplied it manifold, I do not see that there can be grounds for

<sup>1</sup> Doyères found that dried Tardigrades were not destroyed till the temperature reached 140°.

Payen showed that the spores of *oidium aurantiacum* did not lose their germinating power till the temperature reached 140° C.

Milne-Edwards has found that dried organisms could resist a very high temperature.

Instances of great resisting power of seeds to heat have been brought forward by Tyndall and others. Mr. James Sanderson, of Galashiels, tells me that in some specimens of wool obtained from South America, seeds of *medicago* are present, which develop even after the wool containing them has been dyed—*i.e.* after they have been subjected to prolonged boiling and to the action of various chemical substances.

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any longer retaining the view of spontaneous generation. It would just be as sensible to suppose that in the impregnation of the ovum of higher animals it is not the spermatozoa, but some organic molecule accidentally introduced at the same time which causes the development of the ovum, as to suppose that it is not bacteria or their spores but some organic molecule manufactured in or specially altered by passing through the air which produces bacteria in organic fluids and tissues.

## CHAPTER XI.

## RELATION OF MICRO-ORGANISMS TO FERMENTATION.

Summary of what has preceded with reference to fermentation. Relation between 'vital' and 'chemical' fermentations: theories of fermentation. Liebig's views. Alcoholic fermentation: *Pasteur's experiments and conclusions. Résumé.* Butyric fermentation. Formation of pigment by bacteria *Schroeter: Cohn.* Viscous fermentation. Lactic fermentation: *Pasteur: Lister.* Other fermentations, especially the putrefactive: *Lemaire; Caze-neuve and Liron: Paul Bert.* Conclusions.

WE must now pass on to the relation of these bodies to fermentations, and I will here merely indicate the chief points without entering into a discussion on the subject. And first, I may say that it is now admitted by Dr. Bastian, as well as by other observers, that organisms are present in all fermenting fluids. This statement was formerly denied, on account of the imperfection of the methods of examination.

We have already seen in the first part of this work that the cause of fermentation in organic substances was the entrance into them of solid particles held in suspension in air. We have also seen that the cause of the development of organisms in fluids and tissues was the entrance into them of particles suspended in the air. We also know that in all fermentations organisms are present, and that in the absence of organisms no fermentation occurs. What more likely, then, than that the particles which cause fermentation, and the particles which originate organisms, are one and the same? that in fact the fermentation of a fluid is the result of the growth of organisms in it?

The process in these 'vital' fermentations may be brought into the same category as that in fermentation by the 'unformed' ferments, if we suppose that the immediate cause of the chemical change in the former instance is some chemical

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