Symbiotes or Benevolent Microbes and Vitamines

How They Assist in the Life Processes of Higher Organisms: A New Theory

By May Tevis

THE world of science has recently been startled by a new and revolutionary theory concerning the so-called benevolent microbes. It has long been known, indeed, that the bodies of animals contain not only the pathogenic germs which produce disease, but other microbes which are apparently harmless. A well-known French savant, M. Paul Portier, professor at the Oceanographic Institute, has been studying these non-pathological micro-organisms for some years, and has recently advanced the novel hypothesis that they are not merely non-injurious, but are actually beneficial and indeed essential to the vital processes of higher organisms.

It is M. Portier's belief that symbiosis, *i.e.*, the life in common of two organisms which are mutually beneficial to each other, is a universal phenomenon. The classic example of this sort of communal life is the lichen, since every lichen is not a single plant, as it appears to be, but consists, in reality, of two plants, a fungus and an alga, which live together in harmony. According to M. Portier there are no simple organisms except bacteria, all the higher organisms being in reality twofold, consisting, that is, of the organism itself and of the micro-organisms which are distributed throughout its tissues and play an important and necessary part in the processes of life.

To these minute collaborators in the vital functioning of the organism the name symbiotes has been given. These symbiotes, which are very plastic and very active have been isolated and proven to be capable of life outside their host. While widely distributed throughout the body they are found principally in certain organs, particularly in the pancreas and the reproductive glands; they rarely occur in the liver.

During the last year or so M. Portier has published several memoirs concerning his studies of symbiotes in the *Comptes Rendus* of the French Academy of Sciences and elsewhere. Furthermore he has just published a volume entitled *Les Symbiotes* (The Symbiotes), in which he gives voluminous data concerning his experiments with a large variety of animals, ranging from insects to vertebrates, along this line. We are greatly indebted for the information in the present article to an admirable review of the latter from the pen of Henry de Varigny, which appeared recently in the *Bibliothèque Universelle* (Lausanne), since the work in question has not yet reached this country.

M. Portier gained his first idea of symbiotes through the study of certain caterpillars of the micro-lepidoptera, such as the genus *Nepticula*. These caterpillars inhabit minute tunnels which they excavate in the thickness of leaves. They are absolutely aseptic and live upon crushed plant cells whose content is accessible to the digestive juices. Other caterpillars, such as that of the *Gracilaria Syringella*, are also aseptic at the beginning of their existence before they desert their tunnel to live outside. Living then upon more resistant cells they crush them imperfectly. But upon quitting their tunnels they become infected and their digestive canal is full of microbes living upon cellulosic debris which they render soluble. Here we have the beginning of a beneficial association since the microbes live upon the cellulose while the insect lives upon the contents of the cells liberated by the dissolving of the cellulose. Again the digestive juice of those insects which live upon wood (xylophagous) is not capable of attacking cellulose, yet the larvae are enabled to develop by means of microbes which live upon wood by means of cytolysis afterwards supporting the life of the larvae. In the caterpillars of *Nonagria* and of *Sesia*, these well-fed microbes penetrate the epithelial cells of the intestines and there become liquefied; there is an intercellular digestion of the microbe by the intestinal cell. Some of them pass into the blood, where they are intercepted and absorbed by the leucocytes or white blood corpuscles, while some of them escape this fate and become encysted in the tissues. They pass of their own accord into the eggs and there multiply, and when the egg develops the larva is already provided with its symbiotic micro-organism. Thus in this case symbiosis is actually hereditary, and this seems to be intentional upon the part of Dame Nature, since the tissues

which contain the most of these auxiliary microbes are the reserves of fat adjoining the reproductive glands. These inclusions exist in all insects and have also been found in all vertebrate animals thus far investigated.

M. Portier has even succeeded in cultivating the symbiotes found in vertebrates, the easiest cultures to make being those from the lower forms of vertebrates. He has established the fact that the symbiotes of birds and mammals require a temperature of about 40°C., while for those of the lower vertebrates 25°C. is sufficient. It has been proved that the culture takes place through the skin which develops upon the surface, but it is evident that the passage of the tissue into the midst of the culture constitutes a critical period for the symbiote which it does not always survive. A curious circumstance is that the best culture of symbiotes is yielded by a tissue which has just passed through a state of intense physiological functioning.

As seen in the culture the symbiotes are highly polymorphous. They are generally in the form of bacteria of medium length, but by varying the culture medium they can be transformed into short bacilli and even into ordinary micrococci or into large spherical micrococci—or again into highly mobile short bacilli or into immobile filaments. All these are reduced to fragments by the presence of antiseptics. Thus we see that they are highly malleable from the morphological point of view.

They are equally malleable likewise from the physiological point of view. The optimum temperature varies according to the origin. When the symbiote is derived from a frog or a toad, 25°C. is a sufficiently high temperature, while from 42°C. to 45°C. is required when it comes from a mammal or a bird. Thus we see that it produces races which are adapted to the heat of the surrounding medium; however, all symbiotes of whatever origin are capable of adapting themselves to both these extremes and even to temperatures of from 50°C. to 60°C. (122°F. to 140°F.).

In spite of this comparatively great resistance to heat, symbiotes perish ordinarily when in a moist medium at less than 100°C. However, they can be trained so as to resist a temperature of 110°C., or even 117°C. In the dry state they will resist 140°C., but at 150°C. they do not survive more than half an hour.

All antiseptics are fatal to them, the amount and time required being variable. If such experiment be arrested before death ensues symbiotes are obtained whose culture is slower and more difficult and an aberration of form is exhibited.

HOW SYMBIOTES ASSIST IN NUTRITION

M. Portier is convinced that symbiotes preside over two sorts of phenomena in the process of nutrition—simplification and destruction on the one hand and building up or synthesis on the other. When furnished carbohydrates they render them very oxidizable reducing Fehling's liquid just as the organism does. They transform glycerine into dioxycetone; they seem, indeed, to transform proteic substances into acid amines in the same manner as does the organism. From the synthetic point of view they also behave like the organism, since when furnished with saccharose and a nitrate they manufacture a polysaccharid which is very similar to glycogen. As a matter of fact, according to M. Portier, it is really the symbiotes which accomplish this work and not the organism itself.

So far as is now known the symbiotes probably enter the organism with food by way of the alimentary canal. When inoculated experimentally subcutaneously or into the circulatory system they are entirely innocuous, producing no reaction.

One of the most important features of the author's theory is his identification of the symbiotes with the elements of the cell known as mitochondria. These are minute bodies which exist in all cells whether of animals or plants except in most of the bacteria. They are also known as bioplasts, leucites, vesicles, etc. They are usually in the form of small spherical bodies, but sometimes occur as filaments composed of grains, or even as rod-like bodies. M. Portier is convinced that these mitochondria, a definite num-

ber of which exist in each cell, are really symbiotes.

Various investigators have differed greatly as to the function of the mitochondria. Thus Guillemond has shown that they elaborate starch; Policard, that they produce hemoglobin; Mullen, that they make pigment, etc. M. Portier harmonizes these discrepancies by declaring that the mitochondria are really symbiotes—-independent organisms feasting at the same table with higher organisms. The symbiote, in other words, is a highly polymorphous form of bacteria.

Certain histologists raise the objection that the mitochondria are not always capable of being cultivated *in vitro*. The author of this theory answers the said objection in a highly ingenious and interesting manner. He observes that there is no more reason why the mitochondria should be able to reproduce themselves at all times than for men or animals to possess the same prerogative. In men and animals old age causes the loss of reproductive powers, while in the mitochondria the same result is occasioned by the fulfilment of their physiologic rôle. When a mitochondria elaborates any product whatever it becomes a sac containing the said product. In this condition the mitochondria loses its power to reproduce itself by division and remains a mere envelop. The mitochondria which reproduces itself is the one which has not yet become specialized.

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Vital operations are of two sorts—functional activity involving the destruction of reserve material and the liberation of energy and functional repose during which the reserves are elaborated by means of synthesis; as Claude Bernard has strikingly expressed it: "Life is creation." Both these phenomena occur alternately in the same cell and in the same symbiote according to M. Portier—but the symbiote not only manufactures a food reserve but is totally transformed into such an aliment and is entirely consumed at the proper time. The symbiotes share with their sister bacteria an enormous power of synthesis of the most complex and various substances. Bacteria are even capable of manufacturing protoplasm, albumenoid matter from simple inorganic substances, while, on the other hand, there seems to be no organic compound in nature which cannot be disintegrated into its elements by some sort of bacteria.

M. Portier's view as to the part played by symbiotes can be best understood by studying concrete examples. There are certain living creatures, namely, the bacteria, which are sufficient to themselves in the matter of nutrition. Hence they may be termed *autotrophic*; these possess neither symbiotes nor mitochondria—they are themselves symbiotes or mitochondria and derive from inorganic matter the material from which to manufacture organic matter, namely their own protoplasm. All other living creatures, whether plants or animals, contain symbiotes and are, therefore, termed *heterotrophic*. With the exception therefore of bacteria all living organisms present a condition of symbiosis.

What then is the rôle of the cell? In the first place it appears to limit the number of symbiotes contained. Furthermore, it appears to constitute by means of its cytoplasm and its nucleus the chemical milieu which surrounds the symbiote and from whence the latter extracts the raw material required for its activities.

SYMBIOTES AND VITAMINES

A highly interesting point with respect to vitamins is raised by M. Portier, which will undoubtedly rouse wide interest and possible controversy among men of science. It is only a few years ago that the well-known English scientist, Caspar Funk, made the startling announcement that certain wasting diseases, such as scurvy, beri-beri, etc., are really maladies of malnutrition and are due to the lack of certain indispensable elements in the food of the victims of such diseases. These elements were termed *vitamines* by Funk. These vitamins are supposed to exist in certain portions of fruit, seeds and vegetables more than in others. Thus they are contained in the outer coatings of rice, wheat and other cereals and for this reason polished rice and white flour (made from the inner part of the wheat) are imperfect

food, incapable of supporting life unless supplemented by other foods containing the vitamins, i.e., certain acid amines which are indispensable to life, of which the former have been deprived. But Portier holds the opinion that such maladies are really due to a deficiency of symbiotes, or what he calls a state of *a-symbiosis*. According to Funk the sterilization of foods, as in canned vegetables, kills the vitamins, and for this reason such foods tend to produce scurvy. But according to Portier sterilization kills the symbiotes. He has proved by experiments that the latter perish at a temperature of about 120°C. and he points out that this is the very temperature at which vitamins are supposed to be destroyed, remarking that it is much easier to believe in the death of the living organisms known as symbiotes than in the disintegration of a vitamin, which as a matter of fact has been proved not to dissolve until the temperature of 233°C. is attained.

One of Funk's most convincing experiments was the feeding of mice with bread which had been previously treated with alcohol. When fed upon this bread alone the mice soon died, but when the alcohol in which the bread had been soaked was added to the bread the mice continued to thrive. Funk explains this by saying that the alcohol has absorbed the vitamins and carried them away with it. Portier declares in his turn that the alcohol has removed the symbiotes. The latter are found abundantly in bran, and if the bran be treated with alcohol and the alcohol then evaporated the residual liquid will produce an abundant culture of symbiotes. Symbiotes resist alcohol and also resist heat up to 120°C. One of Funk's most telling arguments is that the decortication of rice and other cereals occasions phenomena of malnutrition, which he ascribes as we have said, to the removal of the vitamins, but Portier claims that the phenomenon is due to a lack of symbiotes. Living symbiotes are found in bran and in the integuments of seeds in general. But in the seed they have already been transformed into sacs containing reserve products, and if an animal is given cereals deprived of the coatings of the grain, it is given a food lacking in symbiotes.

But Funk was able to restore pigeons suffering from polyneuritis through de-vitaminized food by administering to them vitamins in the form of a crystallized substance and this substance contains no symbiotes. Portier, however, responds gallantly to this argument by saying that doubtless the vitamin is one of the products of the activity of the symbiote. The same restorative result can be obtained, therefore, by administering either vitamins or symbiotes to the victim of malnutrition. As a matter of fact this view appears to be supported by a striking experiment made by Portier and Bierry. They injected living symbiotes into animals in a state of *a-vitaminosis*, i.e., almost at the point of death through malnutrition. The animals were immediately resurrected exactly as when vitamins are injected.

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It has long been acknowledged that certain plants such as the leguminosae possess symbiotic bacteria and that these assist them in obtaining their proper nutrition. It is but a step further, therefore, to suppose that analogous bacteria, or symbiotes, may have a like function in the nutrition of animals. It is supposed that the symbiotes enter the alimentary canal of men and animals together with their food and the question then arises as to the method by which they become distributed in the various organs where they are found. M. Portier's theory is to the effect that this distribution occurs by means of the blood. It has been shown in recent years that besides the red corpuscles and the white cells, or leucocytes, the blood contains certain minute bodies termed globulins, whose origin and function has never been ascertained. These globulins exhibit a striking resemblance to bacteria. They are in the shape of rods of different sizes, as if some of them were older than others. These rods readily assume the rounded coccus form quite like the mitochondria; they are most numerous in those creatures whose metabolism is most intense. They are markedly diminished in a state of fasting. Portier has conceived the idea that these globulins are really symbiotes employed in the nutrition of the animal.

Granted that the symbiotes are concerned in nutrition the question naturally arises as to whether they play their part in reproduction. A distinguished histologist, Fauré Fremiet, has shown that the ovule is almost entirely lacking in mitochondria while the corresponding male element is well supplied with them. Portier explains this by saying that the mito-

chondria or symbiotes in the ovule have already undergone a transformation into reserve materials, usually fats, as described above. It will be remembered that there are numerous cases of natural or artificial parthenogenesis, in which the ovule develops without being fertilized. The development starts from a mass of matter called by histologists the *vitelline body*, which is of mitochondrial origin. But according to Portier this vitelline body consists of a cluster of symbiotes which have become isolated but are not yet transformed into sacs of reserve materials. These symbiotes when set free by a process which remains to be discovered are capable of producing the development of the ovule in spite of the absence of male element which is so well supplied with symbiotes. It will be seen that this hypothesis supplies an explanation not only for natural, but also for artificial or experimental parthenogenesis, if we suppose that the ovule contains a sufficient number of non-transformed symbiotes to replace those which are habitually introduced by the male element. If this be true the experimenter need only arouse by some suitable agent the symbiotes which exist therein in a quiescent state, or else to inoculate the ovule with fresh symbiotes if those already existing therein are exhausted.

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One advantage of the theory of symbiotes, according to its author, is that it agrees admirably with the "Unicist" theory of life. The view was held by Liebig and J. B. Dumas that the nutrition of animals differs in nature from that of plants, the latter making use of immediate principles by means of synthesis and reduction while animals destroy and oxidize the materials elaborated by vegetable life. In other words the animal merely makes use of building blocks which have been constructed by plants from inorganic matter. This theory was vigorously combated by Claude Bernard, in whose opinion analysis and synthesis are inseparable—the two phenomena being co-existent in all living organisms. This view is termed the Unicist theory. The intervention of symbiotes, "which are derived from the same stock and which are, perhaps, identical, or which at any rate consist of very similar species in all living creatures," says Portier, well accords with the views of the great physiologist.

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If symbiotes wear out and grow old through their activities and their transformations and if they are indispensable to the organism, it is evident that the latter cannot survive unless the supply of young symbiotes is constantly renewed from without; hence if the food fails to contain the necessary supply of symbiotes the organism is in jeopardy, a view which accords with what has already been said concerning vitamins and symbiotes.

Animals fed upon aseptic food which has not been heated thrive perfectly; they also thrive upon food which has been slightly heated, even when the heating has been repeated several times, provided the temperature is not too high. But as soon as the temperature of boiling water is reached some foods become injurious, while all are harmful after being heated to 120°C. (248°F.). In other words a food which has been certainly sterilized by heat is a food which may still retain its physiological, but not its biological value. M. Portier believes that it is the destruction of the symbiotes which has thus depreciated the value of the food. The aseptic life is possible when symbiotes are present, as in the case of the tunnel-making caterpillars referred to above, but is impossible without symbiotes. But it must not be forgotten that the symbiotic condition is a state of normal physiological association, which should not be confused with the septic condition which is a state of abnormal and pathological association.

It has been proved, however, that certain animals exist which are capable of living upon food which is entirely free from symbiotes. Portier explains such apparent exceptions by saying that in all cases where the organism seems to feed upon aliment which is deprived of symbiotes, it is nourished in reality by a "transformator" which intervenes between the feeder and the food—usually by a fungus which contains symbiotes. There are certain coleoptera which live upon the wood of trees in which they excavate tunnels having a characteristic form. Apparently they live upon the wood itself, which is lacking in symbiotes. In reality, however, it is a fungus of the genus *Ambrosia* which lives upon the wood, and this fungus contains symbiotes. The wood which is indigestible for the insect is digested by the fungus, after which the insect feeds upon the transformator

fungus. Another example is of the termite, which stores up in its hill dead wood which it crushes and swallows. It seems to live upon the wood, but this is accomplished in a very curious manner. It collects its own excreta and agglomerates them into spherical granules, forming a sort of cake. When these cakes are examined minutely they are seen to be filled with the mycelium of a fungus which produces spores here and there. In an occupied ant hill only this mycelium is seen, but in a deserted hill there issues from the mycelium a sort of down 4 or 5 mm. in height. The fact is that the ants graze, so to speak, upon this growth, and thus keep it cut short. In reality, therefore, they swallow the wood merely to prepare a suitable soil for the fungus, which is their actual food supply.

Even more curious is the case of the wood-eating caterpillars of the genera *Cossus*, *Sesia*, *Nonagria*, *Zeuzera*. These do devour wood indeed, but when the ligneous paste in the alimentary canal is examined it is found to be filled with micro-organisms, particularly with the conidia of a fungus of the genus *Isaria*, and it is the spores of this fungus which form the food of the insects. These spores, which contain symbiotes, are absorbed by the epithelial cells and the leucocytes. There are even cases in which the intermediary fungus is cultivated neither outside the animal nor in the alimentary canal, but in its very tissues. An example of this is the ordinary aphid or plant louse. This seems to suck the juices of the plant which it pricks. However, it sucks them in through a sort of filter, through which it is impossible for symbiotes to pass. But M. Portier explains that the aphid possesses two lumps of a brown or green substance at one side of the intestine, which are filled with bacterioid bodies which form a mycetome, i.e., a mass of yeast cells. The nature of this green mass has been much debated. Portier considers it merely a collection of symbiotes—a fungus living upon the juices drawn from plants and provided with symbiotes. It is this fungus, he believes, which is the real food of the lice; the latter suck the plant juice to nourish the fungus, which in turn nourishes them.

SYMBIOSIS IN PLANTS

Certain plants also exhibit a state of symbiosis. The seeds of orchids, entirely lacking in the reserves of alimentary substances for the young plant, which most seeds contain, are incapable of germination when sown upon sterile ground, but readily germinate upon soil where orchids have previously been cultivated for the reason that in this case the teguments are invaded by a fungus of the genus *Rhizoctonia*. The cells of the orchid digest the mycelium of the fungus and Portier, therefore, regards the orchid as being epixylous in a manner similar to the cytases which attack the wood: it develops by feeding upon the nutritive matters thus elaborated and it then becomes a source of food supply for the plant, to which it gives over the symbiotes. In the orchid this symbiosis is usually not hereditary, i.e., the symbiote does not exist in the seed as it exists in the egg of xylophagous animals—nature relies upon the symbiotes of the soil to supply the seed. In the *Neottia*, however, the parasitic fungus passes into the seed. Similarly the mycorrhizas of many plants merely represent a case of the symbiosis of a fungus and a plant of higher order.

SYMBIOTES AND DISEASE

It will be remembered that the minute bodies in the blood known as globulins are looked upon by Portier merely as circulating symbiotes. If this be true it is possible that they take part in the defence of the organism through the production of immunity. A number of recent researches has shown that the globulins are probably concerned in the genesis of alexine and of the anti-bodies and also in the elimination of bacteria, especially staphylococci. It has been observed, too, that in a condition of anaphylaxis, i.e., the partial or total incapacity of the organism to react against the introduction of injurious substances, the globulins are almost entirely lacking, and it is suggested that if some means can be found to increase the multiplication of the globulins infection may thus be combated.

We will not here discuss Portier's theory of the possible connection between symbiotes and cancer, since much more work along this line is needed either to substantiate or to disprove his views. In the same way the hypothetical connection between symbiotic action and the sudden variation of species known as *mutation* though provocative of interest can here be merely mentioned.