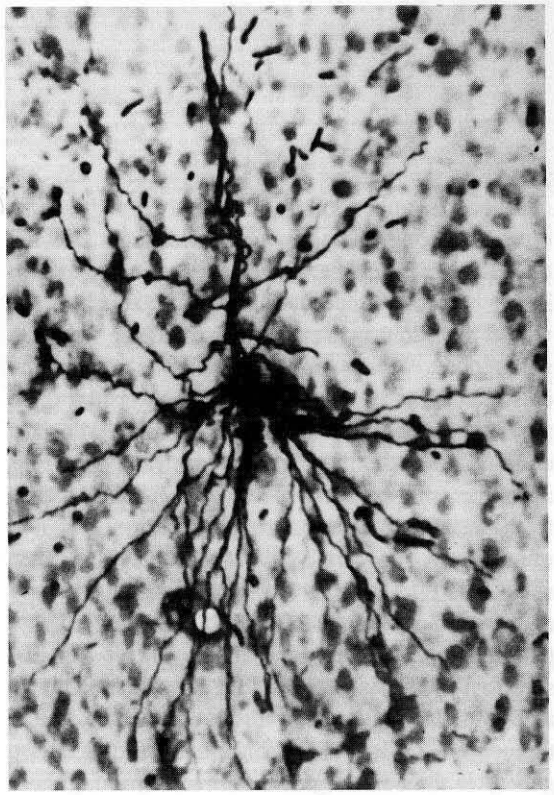
# Nerve, Myelin, and Multiple Sclerosis

*by Jeremy P. Brockes*



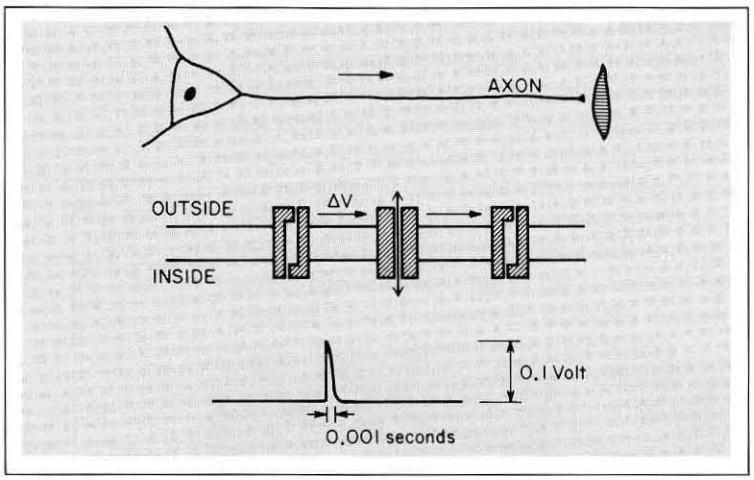
*This nerve cell is seen in a slice or section that has been cut through a region of the brain specialized for vision. The section has been stained in a particular way so that only one of the nerve cells present in the tissue is actually stained. If all of the nerve cells in the section were stained, the whole picture would just look black. (Original photograph by Charles Gilbert and Torsten N. Wiesel.)*

Those who investigate the nervous system may do so at a variety of different levels. The student of behavior may spend much time in the observation of behaving animals. The neurophysiologist who is interested in the mechanisms underlying our senses, such as hearing or vision, concentrates on those areas of the brain that are specialized for these particular functions. In this account I would like to focus on a different level of organization — the cellular one. Just like our other tissues and organs, our brains are made up of cells, and I shall be concerned with various interactions between the different cell types.

Nerve cells are the principal cell type in the nervous system. They are responsible for conducting and processing electrical and chemical signals, and their performance underlies all of those conscious and unconscious activities that we associate with our nervous system. Over half of the bulk of the brain is made up of another cell type called glial cells. Glial cells are unable to conduct and process chemical and electrical signals, but they perform a variety of important functions that support the activities of nerve cells. In our central nervous system (the brain and spinal cord), the principal glial cells are the oligodendrocyte and the astrocyte. In our peripheral nervous system (those parts of our nervous system that lie outside the brain and spinal cord — for example, the nerves passing from the spinal cord to skeletal muscles), the principal glial cell is called the Schwann cell. It is named after a great German anatomist who was active in the first half of the 19th century and was in fact one of those who were first responsible for proposing that biological tissue is made up of cells.

There are many different types of nerve cell in our nervous system. In fact it is a significant point of uncertainty that we do not know how many. Everyone would probably agree that there are at least 100 categories of nerve cell in the brain and spinal cord. Others would argue that there are likely to be thousands, or still more if you looked with methods of adequate discrimination. Since there are well over a million million nerve cells in our nervous system, there is considerable scope for uncertainty on this question.

Nerve cells have a thickened central portion called the cell body. The cell body contains within it the nucleus, which is responsible for directing the construction of all of the different components that make up the nerve cell. Radiating out from the cell body are a number of very fine processes, and it is these processes which are responsible for making connections with other nerve cells in the brain. The fundamental mechanism of communication over long distances in the nervous system is electrical. Nerve cell bodies and their processes serve as conducting elements for tiny electric currents which flow between areas of high and low voltage.

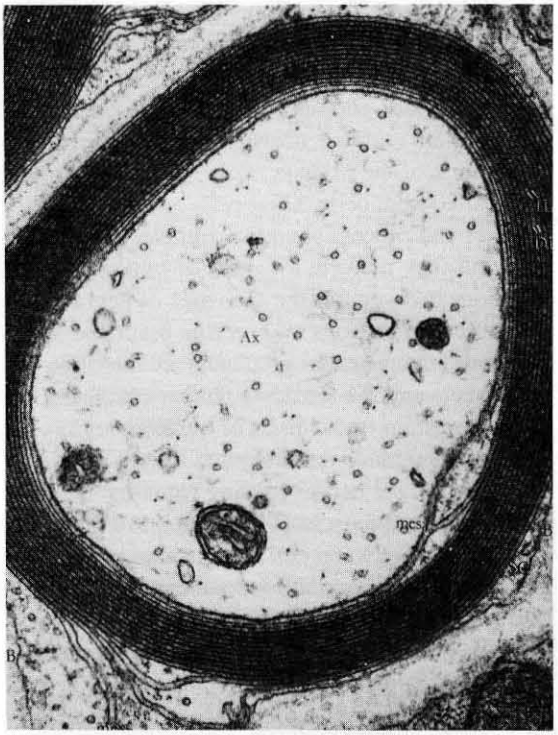



*The top diagram depicts a nerve cell (left) with its axon contacting a muscle fiber (right), while in the center is a schematic drawing of the axon membrane with its voltage-sensitive gates. In its normal resting state (left) the gate is closed, but a change in voltage opens it (center) and permits charged substances to cross the membrane, which changes the voltage and opens the next gate and so on. The general profile of this nerve impulse is shown below.*

The diagram above shows a nerve cell whose cell body is embedded in the spinal cord. It sends out a very long process which makes contact with a skeletal muscle fiber. If this were a muscle fiber in our toes, the process — called an axon — would have to go several meters distance from the spinal cord. When we want to activate our muscles, electrical signals arise from the vicinity of the cell body and travel along the axon leading to contraction of the muscle. The electrical signaling in the nervous system differs in an important way from that which we are familiar with in telecommunications. When we pick up the telephone and speak into it, a train of electrical signals is set up and carried away by cables. The telephone company exerts considerable ingenuity in trying to ensure that the signals are not degraded or attenuated in their long passage through the cables. But the nervous system uses a different principle. It generates the basic signal continuously along the length of the conducting processes. In that respect it can be likened to lighting a fuse of gunpowder.

The axon is a long cylinder covered on its outer surface by a very thin membrane, which serves to separate the outside from the inside. Stuck in and all over the membrane is a very remarkable component which acts like a channel or gate. In its normal resting state this gate prevents the passage of substances across the membrane. The remarkable feature of the gate is that it is sensitive to voltage. When the voltage across the membrane changes, then the gate opens, allowing certain charged substances to cross the membrane, and thereby further changing the voltage across it. The gate soon closes, however, and returns to its initial state. If we look at a little patch of membrane during this process and measure the voltage across it, there is a deflection as the gate opens, and then the signal comes down again as the gate closes. The size of this deflection is about 0.1 of a volt, and it is over in about one millisecond. It is a very remarkable fuse of gunpowder that is capable of being relit hundreds of times a second, since when the signal returns to baseline the axon is able to "fire" many more of these signals. Now it is very important for many of our axons to conduct these signals at high velocities of a hundred or more meters per second. It is usual at this point to remark upon the practical problems that giraffes face in communicating from their spinal cord to their feet. In fact there are many locations in our brain and spinal cord, as well as in our peripheral nervous system, where it is important to have rapid conduction of the nerve impulse. In order to do this, the axons have to be insulated to prevent dissipation of the currents.

The electron micrograph below shows a section that is cut across one of the axons that are specialized for very rapid conduction of the nerve impulse. Because the axon is a cylinder, the section has a circular profile, and it is enormously magnified since its diameter is only 0.01 millimeters. There is a central circular area that is surrounded by about 20 closely packed concentric black lines. The innermost black line is the membrane that we have just been considering; it is the structure which carries the voltage-sensitive components that generate the nerve impulse. Surrounding the cylinder are more of these black lines, and this packed sheath of membranes is the insulating




*In this electron micrograph of a cross section of a myelinated axon, the innermost black line is the membrane in which the nerve impulse is generated. The surrounding lines are the insulating myelin sheath.*

material used in both the central and peripheral nervous system. This material is called myelin, and in the brain and spinal cord it is made by the oligodendrocyte, one of the glial cells mentioned earlier. In the peripheral nervous system, myelin is laid down by the Schwann cell.

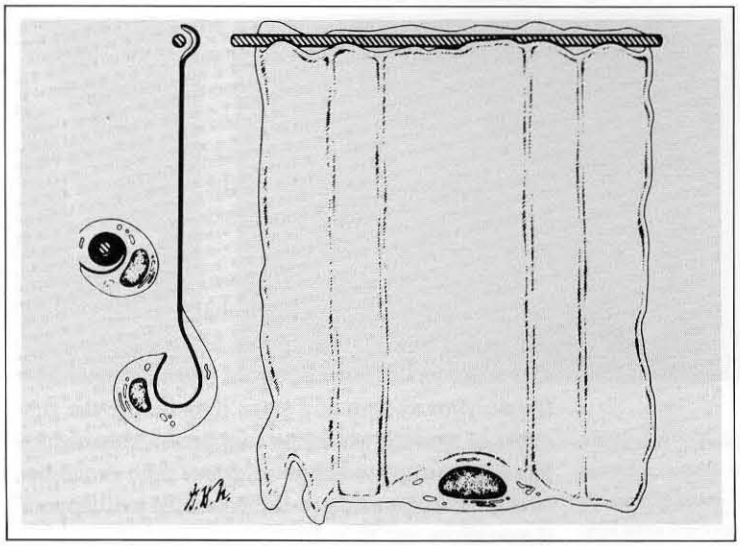
An enormous amount of our brain is given over to myelin, and there is a fundamental distinction between "white matter," containing myelinated processes, and "grey matter," containing the nerve cell bodies. The myelin present in our nervous system testifies to the importance of rapid channels of communication. For a long time it was unclear how myelin was laid down around a nerve, but eventually it was shown that this concentric arrangement is a closely packed spiral, which unwinds as an enormous sheet of membrane.

The questions about how the oligodendrocyte and the Schwann cell interact with the nerve cell have many fascinating aspects to them, but I have chosen to emphasize here the great clinical importance of myelin. There are a number of neurological disorders that result from the breakdown of myelin either in the central nervous system or the peripheral nervous system, leading to failure of conduction of the nerve impulse. The most important and best known of these diseases is multiple sclerosis, or MS, which in many countries is the most prevalent neurological disorder among young adults. In MS the myelin in the central nervous system is lost in certain areas. If we were to look at a section of the spinal cord that had been stained for the presence of myelin, there would be rather discrete patches that did not stain, and where the myelin had been destroyed. These areas are called plaques — a term introduced by the great French neurologist Charcot, who first systematically described the disease over a hundred years ago at the Hospital of the Sâlpetrière in Paris. In the vicinity of the plaque the oligodendrocytes are destroyed and the myelin is lost. Interestingly, multiple sclerosis only affects myelin in the central nervous system. The myelin in the peripheral nervous system, which is the product of the Schwann cell, is not affected. In the vicinity of the plaque the astrocyte, which is the other principal glial cell in the central nervous system, increases in number and takes over the location of the oligodendrocyte. The astrocyte, however, is not able to make myelin so it does not remedy the defect in insulation. One of the remarkable things about the MS plaque is that the nerve axons remain intact despite this great cellular destruction and reorganization.

MS is in many respects a bewildering disease, but most people would agree about its basic qualities. It is not an inherited disease in the strict sense like, for example, hemophilia or Duchenne muscular dystrophy. Certain aspects of susceptibility, however, may well be inherited. The most common age of onset of the symptoms of MS is in the early 30s, but it may be considerably later. The symptoms are highly variable, presumably reflecting in part the location of the demyelinated plaques, though they may affect a variety of neurological functions. But one of the curious features of MS is familiar to almost all patients — the episodes of remission and relapse. A period of remission is one where the initial symptoms either disappear completely or abate for quite a long period, only to be followed by a subsequent period of relapse when the symptoms return.

How common is MS? When we are talking about short-lived diseases, like influenza, it makes sense to talk about incidence — the number of cases that occur in a given year. But when we are considering a long-lived disease like MS, the more relevant figure is the prevalence, which is usually expressed as the number of cases on a given day per hundred thousand of the population. One of the interesting features of MS is the rather marked variation in prevalence with geographical latitude. Although there are some exceptions, it is a rare disease in the tropics. In northwest Europe, the northern United States, in southern Australia and New Zealand, the prevalence may well be at the level of 40 cases per 100,000 or even higher. In the Orkney and Shetland Islands off the north coast of Scotland, the prevalence is a staggering 200 or 300 cases per 100,000. These numbers tell us that many individuals in society suffer from the disease, but it is the celebrities with MS who most effectively draw attention to it. One of the hostages in Iran

*Reprinted from an article by H. deF. Webster in the Journal of Cell Biology 48 (1971).*



*At the far left is a schematic diagram of a profile of a myelinated axon in its normal wound configuration, similar to the micrograph on the previous page. Next to it on the right are two views of the axon and glial cell after the myelin sheath has been unwound.*

was allowed to return home early because he was afflicted with MS. So far as I am aware, however, the most famous patient with MS is the virtuoso English cellist Jacqueline du Pré, whose career was ended by the disease a few years ago, but who has continued to speak out very rationally and courageously for the need for more public awareness of the disease and for increased support of research.

The cause of MS is unknown, and there is no certain cure. One suggestion that I would like to consider a little further is that MS occurs as a result of an environmental agent that impinges on the nervous system at a stage long before the first onset of symptoms. Some support for this idea has come from studies of migrants who move between different areas of the world. What happens if you grow up in an area of characteristically high frequency, like northern Europe or the northern United States, and then move to an area that has a characteristically low frequency, such as South Africa or Hawaii? The general trend in these data is quite interesting. If you move at an early enough age from your residence of high MS frequency, then you can move to your new location and take up its characteristic low frequency. If, however, you move past a certain age from your original residence of high frequency, then it is too late, and you express the frequency of your original residence even though you now live in one of low frequency. The age at which this changeover occurs has not been established precisely, but many people consider that it is around 15. For example, there have been studies of migrants from continental United States to Hawaii, which is a characteristically low frequency area. If they migrate below the age of 15, they express a prevalence that is five times lower than those who move over the age of 15. There is also a considerable amount of data on people who moved from northern Europe to South Africa that in general supports this picture.

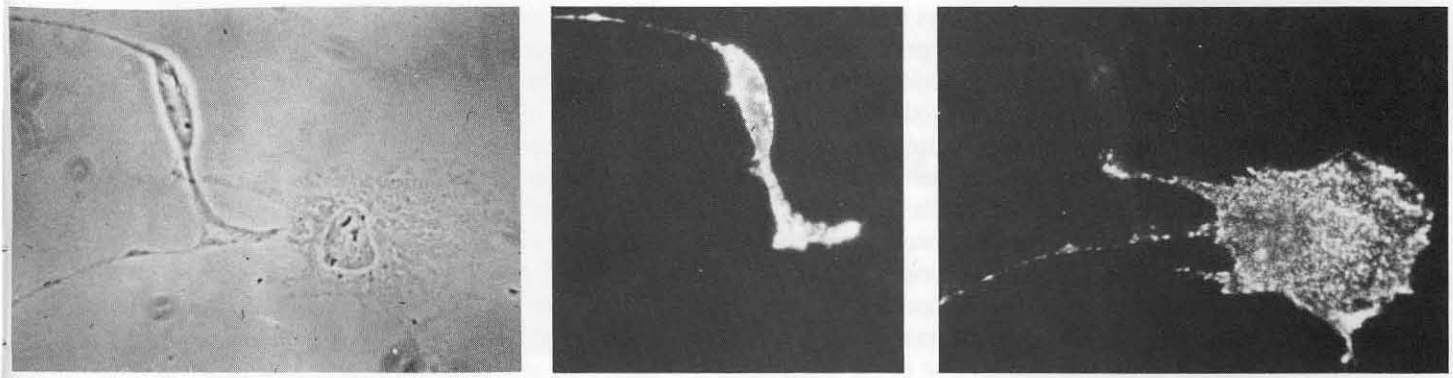
So what does this mean? We do not understand in detail, but some have suggested that at a critical early stage in adolescence an infectious agent, for example a virus, may enter the nervous system. The infectious agent may lie low in the nervous system for ten years or more and only express itself at a much later stage when the first symptoms are evident. The problem with this is that the only decisive proof of such a hypothesis would be to actually isolate and characterize the agent that is being invoked. Unfortunately, all efforts so far to isolate such agents have either been unsuccessful or have not been reproduced in other labs.

There is a second very important aspect of understanding MS, and that is the role of the immune system. When we inject a mouse (the immunologists' favorite animal) with a foreign substance, the mouse's immune system recognizes it as foreign. The immune system responds in a characteristic way, so that if a week later we take a small sample of the blood from this mouse, we can detect components called antibodies. The antibodies bind tightly to the substance, the antigen, that was injected into the mouse. If we look in the white cells of the blood, we can detect cells which again specifically recognize the original antigen. The immune system is a very important aspect of our defense mechanisms to infection, but it is a prerequisite of a system of this sort that it should not recognize the mouse's own components as foreign. If it did so then the mouse's immune system would attack its own tissues with disastrous consequences. There is a set of diseases, called autoimmune diseases, where this happens; that is, our immune system recognizes our own components as foreign and attacks them, leading to their destruction. The circumstances surrounding the removal of the myelin sheath and the oligodendrocyte in MS strongly suggest some autoimmune involvement. It seems that the patient's own immune system participates in the destruction of myelin in the central nervous system.

How would one put this idea, which invokes a role for the immune system, together with the previous suggestion about an infectious agent? If an infectious agent were to enter the nervous system around adolescence, to lie low for about ten years or more, and then put its head above water by expressing itself in the oligodendrocyte, then the immune system might attack the oligodendrocyte and the myelin sheath because it recognizes them as foreign. This is a hypothesis that quite a number of people find attractive, but it is not yet established in any crucial respects. In particular, the identity of the infectious agent that is postulated in such theories is as yet unknown.

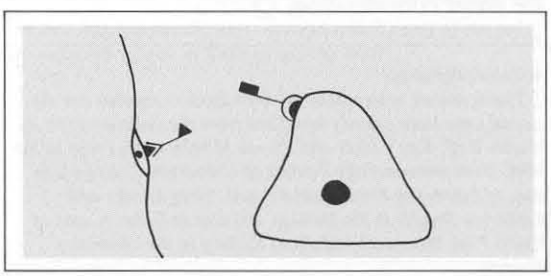
It is often very difficult in medicine to achieve a complete chain of causation, that is, to completely understand what gives rise to a particular disease state. In fact, there are several examples where diseases have been cured without knowing such a complete chain of causation. One example is diabetes, which has been effectively treated by modern medicine without understanding the fundamental defect in the pancreas. In the case of MS, the presence of the intact axons in the vicinity of the MS plaque encourages us to think that it would be good to try to promote remyelination in order to reinsulate the axons. Alternatively, we might try to find circumstances that would enable

*Reprinted from an article by J.P. Brockes, K.L. Fields, M.C. Raff in Nature, vol. 266 (1977).*



the axons to recover the conduction of the nerve impulse. One problem with such aspirations is our ignorance of the basic cell biology of the elements that make up the MS plaque — the oligodendrocyte, the astrocyte, and the nerve cell. The complex architecture of the brain makes it very difficult to understand the elementary interactions between cells which give rise to that architecture. A variety of experience has told us that in such situations where we are faced with a complex biological organization, the most powerful analytical approach is to dissociate the tissue into its component cells, and to take these cells, separate them, and grow them outside the body in cell culture in purified populations. We can then study their properties in defined conditions both when they grow in isolation and in combination with one another. This has been a powerful approach to understanding the elementary cellular interactions that generate complex tissues and organs.

When this is attempted with cells from the nervous system, it is difficult both to identify the cells once they have been dissociated and also to purify them. During the last few years, we and others have been using immunological methods to achieve this end. Although the immune system is a problem in the context of autoimmune disease, it can also be considered in a more beneficent mould — that of giving us substances that we can use to identify and purify cells. The principle of this is very simple. The picture below shows two cells that are different, and one of the ways in which they differ is that they are made of different components shown here by a triangle and a semicircle. We are particularly interested in components like these that lie on the surface of the cells. What we can do is to inject these cells into animals, and the animals' immune system will recognize the triangle or the semicircle as foreign and will make antibodies that bind specifically to them. The antibodies are represented as binding to the two components. We can label these antibodies with different dyes so that they can be distinguished when they are irradiated with ultraviolet light. So what we are doing, in essence, is to use the great discrimination of the immune system to make the distinction between different cell types.



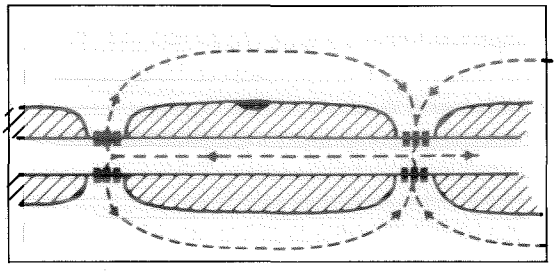
Given that we are able to use antibodies in this way to identify and purify cells from the nervous system, what can we do with the populations that result? The cell type with which we have had most success is the Schwann cell, and the availability of pure populations of these cells has provided a number of new opportunities. One focus of our efforts has been to investigate the process of myelination in a more analytical way. We are now able to separate the two cells that give rise to the myelin sheath — the nerve and the glial cell. They can be studied in isolation and together in culture. One of the insights from this work is that the interaction between the two cells is more complicated than we might have thought. The Schwann cell is not only induced by the nerve to spiral around the axon; it also does not make the characteristic components of myelin unless instructed to do so by the nerve. We are trying to identify the signals that pass from the nerve cell to the glial cell and induce it to do this. The ability to separate the two cells and put them back together again under controlled circumstances may allow us to progress with this difficult problem.

Control of cell division is another aspect of our work. Cells increase in number by doubling certain of their critical constituents and then dividing

***These** two cells from a rat's peripheral nerve are shown growing in tissue culture. The cells have been reacted with fluorescent antibodies and are seen at left under visible light. Under ultraviolet light the fluorescent antibodies can be distinguished, and the spindle-shaped Schwann cell (center) would appear red, while the other cell (right), which is not derived from the nervous system, would appear green.*

in two. Cell division is obviously very important, and there are a number of ways that it is controlled in the body. When we grow the purified Schwann cells, they do not divide even under conditions which allow many other cell types to do so. There is a component present in the brain and in the pituitary gland at the base of the brain which makes the Schwann cells divide. We have been able to purify this component from the pituitary gland and to grow purified Schwann cells in its presence. This component also acts on astrocytes, the glial cells that proliferate in the vicinity of the MS plaque. One of the interesting things about it is that it seems to be a product of nerve cells. Nerve cells are distinguished by the fact that they do not divide, which is one reason why neurodegenerative diseases are so serious. Yet we have found that nerve cells apparently make a component which is able to stimulate other cells to divide. There is considerable indirect evidence that nerve cells play an important role in regeneration in a variety of contexts. The fact that we have been able to get hold of a defined component of this sort allows us for the first time to investigate some of these issues directly.

Another focus of our research is the voltage-sensitive component mentioned earlier that is located in the nerve cell membrane and is responsible for generating the nerve impulse. In the diagram of a myelinated nerve process below, the myelin is represented by cross-hatching, and one segment is laid down by each glial cell. Before we reach the next segment there is an area of bare nerve membrane, which is called the node. It has been found in the last few years that essentially all of the voltage-sensitive components that generate the impulse are located in this bare membrane between adjacent myelinating glial cells. None of them are found in the nerve membrane underlying the glial cell. This is something that makes very good sense for rapid conduction in a myelinated axon. When the axon is demyelinated, however, the distribution is inappropriate, and this is the reason why conduction fails. It would be much better if the distribution were uniform after the myelin was removed. We have very little understanding of the factors that control the presence of this component in the nodal membrane. In the last few years, our colleagues in the chemistry division at Caltech have made considerable progress in removing this component from the membrane and purifying it. During the last 18 months we have joined with them and have been able for the first time to make antibodies that recognize this component. These antibodies should allow us to investigate those factors responsible for localizing the channels in the node. We plan to explore this issue by using the defined culture systems that I discussed earlier. One obvious question that we cannot answer at the moment is whether the glial cell plays a role in segregating the channels to the node or whether the nerve cell is able to put them there in an autonomous way.



*In this diagram of successive segments of myelin laid down around a nerve axon, the myelin is crosshatched and the nucleus of the glial cell is shown as a black semicircle. The voltage-sensitive gates represented by black rectangles are localized at the node between successive segments. The dotted lines and arrow indicate the flow of current through the gates and into the axon.*

All of these issues are relevant to the environment of the MS plaque. We would like to understand how to induce cells to remyelinate, and we would therefore like to identify those signals which the nerve cell normally passes to the glial cell to instruct it to make myelin. We would like to understand the factors controlling cell division in the vicinity of the plaque, and we would also like to promote the redistribution of the voltage-sensitive component that is responsible for the nerve impulse. These problems are best studied in the isolated and defined conditions of cell culture. The availability of purified Schwann cells also raises the long-term possibility of introducing them into the central nervous system to see if they are capable of remyelinating axons that have lost their myelin.

It would be irresponsible to minimize the difficulties associated with diseases like MS or to underestimate in any way the value of direct clinical studies. Nonetheless, at a time when support of basic research is rapidly dwindling, it would be equally irresponsible not to point out that such research may be directly relevant, and often in an unpredictable way, to the clinical problems that are under consideration. □

*Acknowledgments*

*The research topics from my own laboratory that are discussed here have greatly benefited from the collaboration of Martin Raff, Kay Fields and Rhona Mirsky when I was in the MRC Neuroimmunology Project at University College London, of Lawrence Fritz, Karl Fryxell, Greg Lemke and Katherine Stygall in the biology division at Caltech, and of Hsiao-Ping Moore and Michael Raftery in the chemistry division.*