

# EUROBRAIN

## Max Cowan Lecture

### Plasticity in the Nervous System

### A Sea Change: And Why It Has Taken So Long To Accept It

Welcome to the first Max Cowan Lecture, sponsored by the Dana Foundation. Max was a strong supporter of the work of the Dana Foundation, in the United States through the Dana Alliance for Brain Initiatives and also through the European Dana Alliance for the Brain, and was an enormously influential neuroscientist in his own right. It is particularly apt that this first Max Cowan Lecture will be given by Geoffrey Raisman. Geoffrey was brought up in Leeds in the north of England, the son of a tailor from an immigrant family. He came to Oxford in 1956 and was a student of Max Cowan's when Max himself was still a clinical medical student. He did his PhD, a still very well known work on the connections of the hippocampus. He is primarily a neuro-anatomist. He applied electron microscopy, in one of the earlier applications of electron microscopy, to the nervous system and his work on the hippocampus led to the notion of plasticity. He made the first demonstration of the formation of new synaptic connections after injury in the hippocampus working in a laboratory which was directed at that time by the endocrinologist Geoffrey Harris. He made the first demonstration of gender differences in the brain, in the hypothalamus. He went to Harvard in 1968 and worked there for a year. He became interested in the question of repair in the central nervous system and after working on transplantation of embryonic hippocampus came to the same conclusion as Albert Aguayo's group that the key to regeneration of transplantation might lie in the special characteristics of glial cells.

He has been at the National Institute of Medical Research in London since 1976 and has developed a very strong area of work on the influence of olfactory ensheathing cells on regeneration, about which I am sure he will tell us, working over the past twenty years on spinal cord injury and repair. He has effected the first and only demonstration of a repair of breathing with high spinal lesions and has developed this work consistently, right through to the stage of potential clinical trials. He will be moving to the Institute of Neurology in Queen Square in London on 1 October [2004], where he will be moving on to clinical application.

I have known Geoff Raisman for well over thirty years and have enjoyed his company in many areas outside science, discovering the wide range of his knowledge and scholarship. I remember particularly taking a trip in Iran at the time of the Islamic revolution, discovering Geoff's enormous knowledge of the Achaemenid period and the whole of the history of the Middle East. He reads and to some extent writes Chinese and Japanese and is interested in classical Chinese literature. He is the author of a book called *The Undark Sky*, which is a history of his own family's emigration from Lithuania and the complications of gambling in that process of immigration to England.

It is a great pleasure to introduce Geoffrey Raisman to give the first Max Cowan Lecture.

Professor Colin Blakemore

**Geoffrey Raisman applied electron microscopy, in one of the earlier applications of electron microscopy, to the nervous system and his work on the hippocampus led to the notion of plasticity. He made the first demonstration of the formation of new synaptic connections after injury in the hippocampus.**



[1]

I have decided to begin my story with my trunk. This trunk is maybe a hundred years old – even older than me [1]. When my mother bought it, it was very old and battered and cost £ 2 – and that was a small fortune. As a comparison, five pounds was a lot of money for a week's wages at that time. The trunk contained the suit that my father had made for me, and with it I moved from the working class streets of Leeds to Oxford, the incubator and cradle of the ruling class of the British Empire. Up till then I had only shared a bed with my uncle Myer, and now I had a servant to make my bed, to tend the fire and to check that there were no girls in the room at night.

As a medical student, I had two tutors, one hour a week with each, one-to-one. Nothing was too hard for us. My physiology tutor was Percy O'Brien, a fiery little Irishman. As a hobby he had learned Egyptian hieroglyphs. Like a Japanese Buddhist master, he used to make a point by striking his students with his fist in the middle of your chest. My anatomy tutor was Max Cowan. I was a first-year medical student, Max was a clinical student. So it was a case of the senior student teaching the junior. I once said to him how much I appreciated O'Brien. Later I realised that had made him jealous – he wanted to be the one his student appreciated – and he remained jealous of O'Brien all his life, long after O'Brien had gone. I thought it was really wonderful that Max wanted to be the appreciated teacher.

We used to take tutorials in anatomy for one hour, walking round the University Parks, beside the River Cherwell, where the grass grew three feet tall in the water meadows. Max would talk about the connections between the cingulate cortex and the anterior thalamus. One of his sayings that stuck in my memory was: "If you read something carefully and you do not understand it, that probably means it was because the writer didn't understand it."

I started research with Max. I was his first and only PhD student in England. At that time the

interest in neuroanatomy was as to how nerve impulses reached the cortex. Professor Sir Wilfrid Le Gros Clark, who was the head of the department, was interested in how the 150-odd cytoarchitectonic areas described by Brodman were related to the rest of the brain. He was a pioneer in the experimental method of determining connections by making lesions in the cortex and looking for which areas showed retrograde degeneration. In fact, Le Gros Clark was the man who established that the geniculate bodies were part of the thalamus.

With Max, I worked on the connections of the hippocampus, one of the oldest and most mysterious parts of the cortex. We were interested in where the hippocampus got its input from. It is the septum. It is one of the very rare mistakes of Cajal that he did not realise that the septum projected to the hippocampus as well as the other way round.

Max was always interested in new technology. At that time the exciting frontier was electron microscopy. Its application to the nervous system had been pioneered in the late 1950's by Palay, who produced the first anatomical pictures of synapses – the tiny axonal expansions which contained synaptic vesicles and which made contact with postsynaptic elements at synaptic thickenings [2]. It was the method of preparation of the tissue that was so difficult – the stringent requirements for fixation, the rare and dangerous chemicals, such as osmium which had to come from behind the Iron Curtain, the new embedding resins and curing, and cutting sections on knives of broken glass, or precious diamond, and the demanding technology of viewing with an electron beam instead of light.

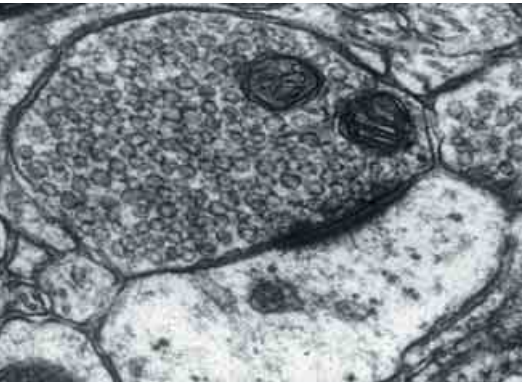
Up to now, we had been using lesions simply as a technical device to enable us to trace nerve fibre connections in the brain. The use of the electron microscope increased the magnification from thousands to millions of times. And it opened up new vistas for studying the effects of lesions. Now instead of simply



Prof. Geoffrey Raisman

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[2] This is a picture of the synapse formed by hippocampal fibre in the septum



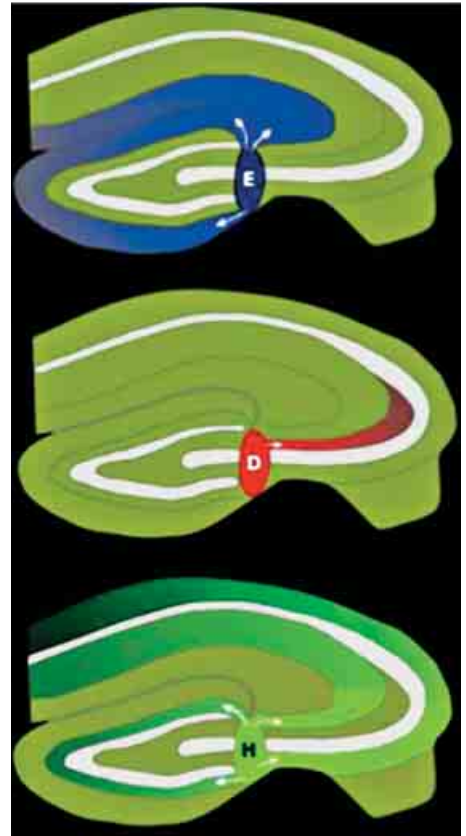
[3]

using lesions to trace pathways, we could use them to study how the brain responds to injury. I remember one evening standing on the corner of South Parks Road with Max. It was getting dark, people were getting on their bicycles. Max was going in one direction home, and I was going in the other. We were standing at the moment of parting. Max was talking about lesions and their effects on synapses, and was pondering the question of what happens when an axon terminal is lost [3].

What provoked that question was a thesis Max was examining. It showed that after injury the synapses at the ends of the cut axons were lost – as expected – but that the synaptic thickenings remained without any axon terminals. They were called 'vacated thickenings' – empty seats in a fully booked auditorium. Max put forward the idea – which I have followed all my life – that maybe after injury new connections form. It was an unbridled piece of heresy. In mediaeval Spain people burnt for less. The idea stayed in the back of my mind, and one afternoon, while the sun lingered over the waving grasses in the University Parks, some time after Max had left Oxford to go to the United States, I was very bored teaching anatomy and used to look for a way to relieve the tedium. I was talking to a student about the nerve supply of some muscle in the arm, or something like that, and I threw up an electron micrograph and said: "What do you think that might mean?"

The micrograph showed an axon terminal making two synaptic contacts at two different synaptic thickenings [4]. "Do you think," I asked "that this could be the basis of memory? That new synapses may be forming in the adult brain?" It was the first time I clearly saw the possibility of what I later came to call "plasticity" (Raisman, 1969).

Through a lot of experiments we were able to demonstrate that what was happening after injury was that, if one synapse was lost, an

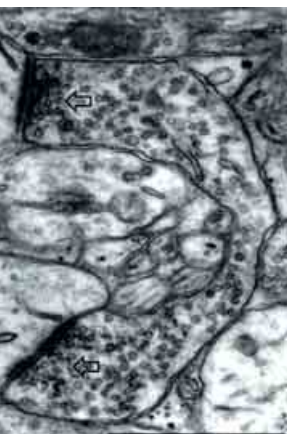


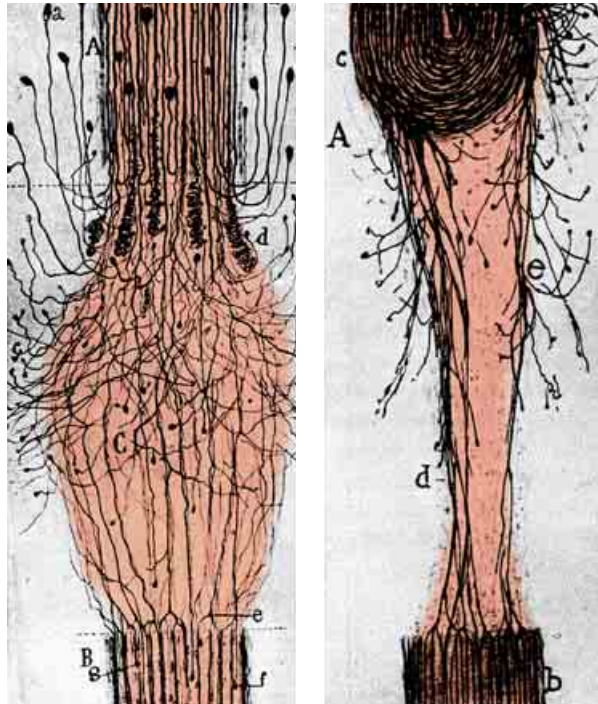
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adjacent terminal sprouted out a new contact to fill the space. If you originally have two different nerve fibres and one is destroyed, then the remaining one sprouts and takes over its territory. What was new was the idea that the adult brain, the adult central nervous system, could do anything after injury.

I wanted to explore the ability of the adult brain to accept new connections, and I used the technique that Anders Björklund was developing at about that time for transplanting embryonic tissue into brain. For our experiments I used a Thy-1.1/Thy-1.2 allelic marking system. The illustration is from a section of adult hippocampus which has received a transplant of a piece of embryonic entorhinal area [5]. It clearly shows a newly formed axonal projection from entorhinal area to hippocampus. We found, using this system, that if you transplant into adult brain new connections form, but the connections depend upon the type of tissue you transplant, so transplants of entorhinal area tissue make one type of pattern, dentate gyrus tissue makes another pattern and hippocampal tissue makes yet another distinctive pattern (Zhou et al., 1985). The point about this experiment is that the adult brain can not only receive new patterns of connection and but can also accurately regulate which pattern it gets. Thus there remain, within the adult brain, both the power to form new synapses and the power to accept new synapses, and that immediately

[4]





[6]

raised the question, after injury, why cannot the originally cut fibres regenerate?

The ability of cut nerve fibres to regenerate had been known for over a hundred years. This is Cajal's picture of crush of the sciatic nerve [6]. You can see that the fibres have advanced beyond the injury, and they have regenerated. We can see them forming growth cones which explore the environment. Some go down into the distal segment of the nerve. Even if you cut and turn the nerve away, fibres still find their way, less efficiently, into the distal stump. In the peripheral nervous system, these growth cones explore the environment and are able to succeed on restoring original connections.

In the central nervous system, the brain and spinal cord, the outcome is quite different. The illustration shows a lesion that we made in the corticospinal tract of the rat [7]. You can see the growth cones. Individual fibres explore the environment, but they are unable to regenerate to their original destinations and unable to restore their lost connections.

[7]



[8]

On a visit to Prague a couple of years ago I took this photograph of the damage caused by the flooding of the river [8]. Here we can see all the chaos of disorganisation. You can see the telephone cables have come out. The people who were at the two ends of those telephones are still there. But the cables can no longer connect their voices. What is needed is the reconnection. What do you need to reconnect those cables? And it was that thought that raised the question of what is

the pathway along which nerve fibres grow. And the rest of my interest is in fact not about the nerve fibres, but about the pathway.

We see the hippocampus, the septum, and the pathway between them, called the fimbria [9]. What I found – or, rather did not find – was that no one had ever taken an interest in what such pathways consisted of. With Mitsuru Suzuki, who was visiting from Japan for a few years, we looked at the pathway and found something that really had not been described before, or had only just been shown in the odd illustration, and that was that the pathway is made up of glial cells arranged in long unicellular rows of indefinite length (Suzuki and Raisman, 1992). This one is 270 cells long, in fact [10]. Within those rows are four different types of cell. Astrocytes, oligodendrocytes and microglia, form an elegant, regular tapestry, a crystalline structure of living cells.

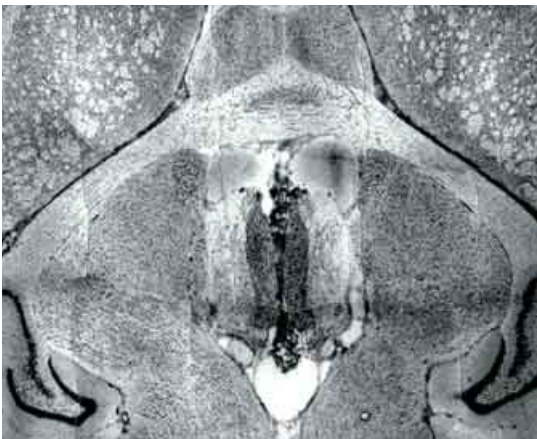
This living tapestry is damaged when nerve fibres are cut. The injury not only severs the nerve fibres, but it damages the pathway, and all these pathway cells respond. How can we reorganise the pathway?

The idea of repairing the central nervous system comes from Cajal, who in turn acknowledges the Italian Rossi, in the 1870's. The idea was: if damage occurs to nerve fibres in the brain or spinal cord, where they cannot regenerate, can we transfer something from peripheral nerves, which can regenerate, into the area of damage? What you can transfer is glial cells. Albert Aguayo and his team showed that, if you cut the optic nerve, you can transplant a piece of peripheral nerve containing Schwann cells (which occur only

[10]



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[9] The accompanying figure shows a section through the brain

in peripheral nerve) and the nerve fibres will grow from the retina across the Schwann cell bridge and make connections. So this cut central nervous pathway can be bridged by a peripheral nerve.

But the bridging effect of transplanted Schwann cells is very inefficient. When a piece of peripheral nerve is transplanted into the central nervous system, cut axons will sprout and grow into the piece of peripheral nerve but very few will leave it. Thus, although axon growth is stimulated, only very few of the sprouting fibres get out of the bridge.

So Schwann cells are good at supporting regeneration in the peripheral nervous system, but poor at allowing re-entry into the central nervous system. Where can we get glial cells that will do it? The answer came from work on the olfactory system, the nerve fibres originating in the nasal lining, and sending their fibres through the skull to make connections with the olfactory bulbs at the front end of the brain. Work with methods of detecting the formation of nerve cells showed that new nerve cells are continually formed in the olfactory system. As a result, the olfactory nerves continually being replaced throughout adult life and the olfactory nerve fibres are continually able to enter the central nervous system. So whatever pathway cells they are using as their pathway have the property of allowing re-entry to the central nervous system. Those cells are called olfactory ensheathing cells.

I am going to talk about examples of two systems in which we have obtained repair of spinal cord by transplantation of olfactory ensheathing cells. The first is the adult rat corticospinal tract (Li et al., 1997). Ying Li, who has been with me for seventeen years now, worked out a technique to make a lesion destroying the corticospinal tract on one side. Into that we inject olfactory ensheathing cells. Rats with these small unilateral lesions localised to the dorsal corticospinal tract look very, very normal and it took us months to find

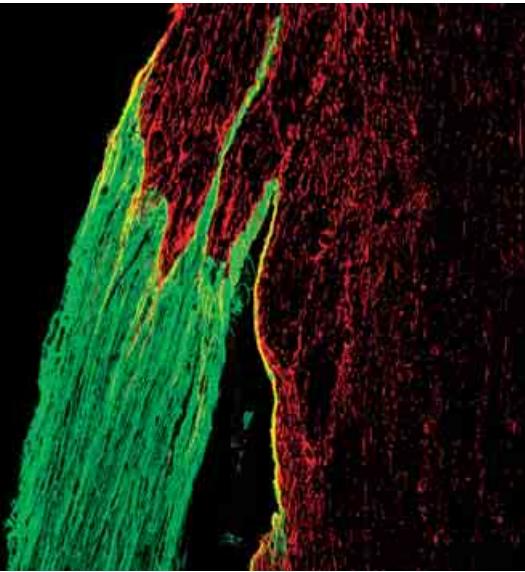
any test. In the end, we modified a test described by Whishaw. The rat comes to the front of the cage, it looks at you with interest, you give it little bits of food, it puts its paw out and takes them. The normal rat will take food with either paw. After a lesion of the corticospinal tract the rat cannot use the paw of that side for retrieval. The paw is not paralysed, it is not unable to move, it can be used in walking, climbing, in manipulating food. What it cannot do is to use it for retrieval. And this loss of function is permanent. After transplantation, however, the function returns.

If we make a much larger lesion and destroy all the tracts in the spinal cord on one side – hemisection – once again within a few days these rats are indistinguishable in their cage behaviour from normal. The functional test is to let the rat climb up a grid. In normal rats all the paws, without fail, grasp the bars. After hemisection the back paws perform normally; the front paw on the unoperated side is also normal. But the forepaw on the operated side misses the bars. It is not paralysed, it is not weakened, it simply cannot find the bar. We transplant cells into the hemisection using a matrix made by the cells themselves in tissue culture. After transplantation there is an enormous improvement (Li et al., 2003).

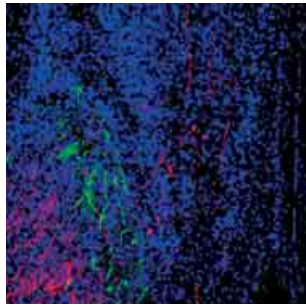
In making a lesion at the upper cervical level of the spinal cord, we also sever the supraspinal nerve fibres from the brain stem respiratory areas coming down to the phrenic motor neurons. We can see the effect by recording from the phrenic nerves. When we look at the hemisectioned rats we get a normal recording on the unoperated side. But on the operated side, the phrenic nerve is silent. Once again after transplantation we find a return of rhythmic activity in the phrenic nerve of the transplanted side.

Our results show that there are cells in the adult olfactory system that can be used to repair damage to the spinal cord and in the

[11]



[12]



future, we hope, in the brain. In the rat we started off getting the cells from the olfactory bulb. But, of course, you can also get them outside the skull from the olfactory mucosa, which is important if we are talking about people because, rather than having to go through the skull and take out an olfactory bulb, which will not regenerate, we can go through the nose, as Mackay-Sim and his colleagues in Brisbane have already done, and take out a patch of olfactory mucosa and grow the cells in culture. The area from which we have taken it will also refill itself from stem cells, because it is a self-reproducing epithelium.

We are planning to try out transplantation of olfactory ensheathing cells, in brachial plexus avulsion, which occurs when people fall out of cars or fall off motor-bikes and hit their shoulder or the side of their head. They pull the roots out of the cord. Brachial plexus avulsion results in a totally paralysed flail arm, with no sensation and often a considerable amount of pain, which can be the main feature. The illustration shows the normal fourth lumbar dorsal root in the rat, and illustrates the point at which the peripheral nerve enters the central nervous system [11]. The junction is where the peripheral nerve tissue stained in green makes contact with the central nervous tissue in red. With Tom Carlstedt, a neurosurgeon who works on repair of brachial plexus avulsion in human, we investigated the effect of cutting the lumbar root and replacing it with olfactory ensheathing cells in the gap. The illustration shows the regenerating nerve fibres going through the graft [12]. They re-enter the spinal cord and run up through the dorsal columns. Our first estimate suggests that as many as 10% of dorsal column fibres regenerate through these grafts (Li et al., 2004).

There have been two strands in this work which Max Cowan started. One strand is the idea of plasticity. Plasticity means more than the simple ability to change, it means the ability to do something positive. Plasticity leads

to hope. The other strand is the idea of humanity. If we get to repair of the spinal cord, if we get even part of the way, or if we fail, even our mistakes might be useful. We are doing something of value to people. Plasticity and humanity are related. Plasticity of the brain has produced our society and it will decide our future. I think we have only glimpsed what plasticity is about. Plasticity is learning. And history is the story of change. History is the story of plasticity.

I remember that I used to meet Max Cowan from time to time coming into meetings like this one. I never made appointments with him – he was far too important for that. He had a voracious appetite for knowledge, for science. He used to sleep two and a half hours a night. I think he read one book per night. Max had started off as a history student in South Africa, I think in Renaissance history. I think his hero would have been Machiavelli. He studied power carefully, and when his turn came, he applied it. He detected trends in neuroscience, such as electron microscopy and methods of tracing connections. He was one of the first to see how molecular biology and development would affect neuroscience. He applied power in running departments, journals, societies, grant bodies. I have to say that, as far as I can see, his judgments were all good, and his effects on our community and on the development of neuroscience were nothing but good. To use a phrase from James Joyce, he was one of the rare people who kept the kettle boiling.

In this part of his life, I was not involved in any way. But I used to meet him coming into meetings like this, going in the morning, and I would sort of detect that his steps were a bit reluctant and hesitant. So were mine, and maybe he saw that. He would say, or I would say, "How about a cup of coffee?". And we would talk about old times and Oxford – he was still jealous of O'Brien – and half a day would pass. Max too was working class. His father was an out-of-work Scottish miner who

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went to South Africa. It was a sort of unspoken bond between us. At times Max seemed to crave company.

As the years went by I didn't see him often. Our lives were so different, and from my corner of some lab I would think of him in the big stage. But from time to time I would write something or get a review and I would think, "What would Max think about this?". One time when I met him I plucked up the courage to say, "Max, I hope one day you will be proud of me". He said, very softly, "Geoff, that would make me happy".

Max's heretical idea about synapse formation in the adult brain after injury had – although neither of us realised it at the time – set the course for my professional life. But heretical ideas were not unusual for us, they were the normal fare of our conversation. The temporary parting on that street corner has now become a permanent one. But sometimes I wish there was a secret street corner, somewhere at dusk, while the people got on their bicycles, and where the scientific conversations of today could be made more heretical, in a world where people did not accept and repeat so readily, and unquestioningly, the ideas handed down to them by those with the scientific power to grant grants, accept papers for journals, release venture capital funds, and make or break careers.

Max came from nothing to a great level of prominence and influence, and it seems so soon that now he is nothing again, as we all shall be. I wondered what all the struggle was

about. But, you know, things were so different then. There was one-to-one teaching, when today there are classes of hundreds. That is the change of plasticity, the history. History has an irony and it has a romance. One of the great English historians explained that the romance of history is to think that in this place where we are now were other people in the past, people who lived quite different lives from ours, who thought quite different thoughts, who had quite different feelings. I would put the same idea another way: history is the contrast between what changes and what does not change. What has changed is the way I was taught as a student. It was mediaeval, and it was wonderful. That will not come again. Max has gone. That has changed. What has not changed is the relationship between human beings. We have not changed. Max was a scientific father to me, and to many others. We can all be scientific fathers, or mothers.

Sometimes when I have taken a student out for a meal, he or she might say, shyly, at the end of it, "Can I pay my bit?" or "Can I help?". Of course, they have no money.

I say, "Yes, certainly you can, and you will. But for now it is a debt. My teachers paid for me. You remember it and carry it with you. You will pay it back." I think that that is a big part of the feeling of what Max passed on, and what we can all pass on.

Professor Geoffrey Raisman  
Lisbon, 13 July 2004

## REFERENCE LIST

**LI Y., CARLSTEDT T., BERTHOLD C.-H., RAISMAN G., 2004**

Interaction of transplanted olfactory ensheathing cells and host astrocytic processes provides a bridge for axons to regenerate across the dorsal root entry zone.

*Exp. Neurol.* 188: 300-308.

**LI Y., DECHERCHI P., RAISMAN G., 2003**

Transplantation of olfactory ensheathing cells into spinal cord lesions restores breathing and climbing.

*J. Neurosci.* 23: 727-731.

**LI Y., FIELD PM., RAISMAN G., 1997**

Repair of adult rat corticospinal tract by transplants of olfactory ensheathing cells.

*Sci.* 277: 2000-2002.

**RAISMAN G., 1969**

Neuronal plasticity in the septal nuclei of the adult rat.

*Brain Res.* 14: 25-48.

**SUZUKI M., RAISMAN G., 1992**

The glial framework of central white matter tracts: Segmented rows of contiguous interfascicular oligodendrocytes and solitary astrocytes give rise to a continuous meshwork of transverse and longitudinal processes in the adult rat fimbria.

*Glia* 6: 222-235.

**ZHOU C.F., RAISMAN G., MORRIS R.J., 1985**

Specific patterns of fibre outgrowth from transplants to host mice hippocampi, shown immunohistochemically by the use of allelic forms of Thy-1.

*Neurosci.* 16: 819-833.



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