Genopole ® : la médecine de demain s'invente aujourd'hui

Mercredi 4 Février 2004

TIM HUNT

Nobel Prize in Physiology or Medicine in 2001

The control of cell proliferation and the problem of cancer

I must apologize because I cannot speak French – once upon a time I knew schoolboy French, but now I do not. I found this slide in a junk shop in Japan and I have asked many hundreds of Japanese people to translate what it says and what they always say is, "Very difficult, writing too fluid…" So if anybody can translate it, I would be very grateful, but I do not hold out much hope.

I start with my daughter Celia and the cat Minky. This cat, although alive at the time of the photograph, is now dead, eaten by a fox. The reason for this next slide is a question that Celia asked at bedtime very shortly after the Nobel Prize in medicine or physiology of 2001 had been announced. Suddenly you go from being an ordinary scientist to some sort of godlike, all-knowing creature, and I think it is important to realize that one actually changes very little. Celia asked why the ceiling is opaque. I had no trouble with that: it absorbs light. I then looked out of her window and wondered how the hell do the photons get through glass? I thought about this and said, well, gas is a liquid and photons get through gases and liquids, except they do not get through mercury. They bounce off mercury, so it has clearly nothing to do with the liquidity of the substance in question.

Then I thought about the element carbon, and I got a little closer to the truth. Black cars use carbon particles to make them black, highly absorbent, but those of you who have rings on your finger with diamonds in them will know that diamond, which is also elemental carbon, transmits light beautifully. That then said that it had something to do with the electronic structure of the substance in question, but I started to ask people. Although I had done physics at school and although we had measured the refractive index of things, nobody had ever asked that simple question, how do the photons actually get through the glass whose refractive index we are measuring? So I found myself finally sitting next to Aaron Klug at a rather grand luncheon party at a British ambassador's residence in Sweden. I said, "Aaron, you know about these things, how do photons get through glass?" He said, "Well, Tim, you really need to understand Schrödinger's equation." At that point, I knew I was lost.

We can find Schrödinger's equation, actually on his tomb. The artist who drew it made a mistake. There is a white dot on the right[2]. It had to be painted out by a well-known physicist who visited the tomb and said, "hey you have got it wrong!"

This is Schrödinger's equation and it looks like a very simple little equation, except that it describes the change in the wave function with respect to time. Of course, one has to consider the wave function in three dimensions and I think this is a just a one-dimensional solution. *i* is of course the square root of -1. Right there you have a really serious difficulty if you are a bad mathematician like me. *h/* is Plank's constant divided by 2p. *Psi dot* is the rate of change of the wave function with respect to time. *H* is the Hamiltonian operator and *psi* is the wave function. Now, what is the Hamiltonian operator?

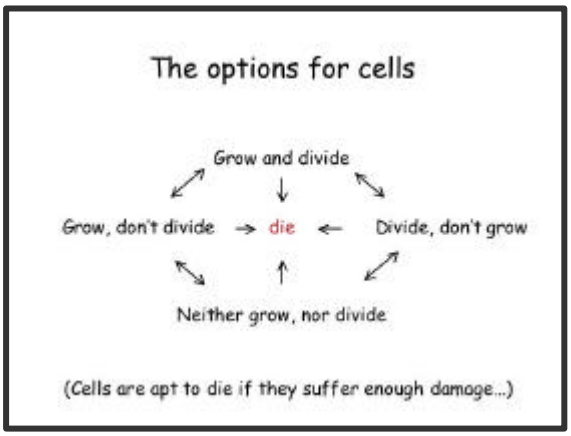
Even the wretched Hamiltonian is not simple. It is not just *H*. It has this amazing symbol, upside-down delta, which I do not even know how you pronounce, let alone what it stands for. Maxwell used this in his famous equations to describe the electromagnetic field. All of this amounts to saying that although people understand very well the mathematical underpinnings of how light gets through stuff, we poor biologists are pretty much at a loss.

Some of you may have read Richard Feynman's excellent, but to me almost wholly incomprehensible, book called *QED* (quantum electrodynamics). This was actually about 1948 and they did not win the prize until 1965. Feynman says this is a theory – the theory of the interaction of photons and electrons – so accurate that if you were to measure the distance from New York to Los Angeles, it would be accurate to within the distance of a human hair. Now, biologists, I would contend, feel quite happy if their experiments are accurate to +/- 10% - a very big difference. So this is a very interesting question about the relationship of physicists and physics and biologists and I can illustrate this very well by another example, which is, I think, the rule by which we operate. Aristotle, who was perhaps the world's first and greatest scientist, had a law of motion. What Aristotle said was that when you push something, it moves, and when you stop pushing it, it stops. Obviously, that is true – I have just shown you. Unfortunately, however, it held up the development of physics by about 2,500 years because Newton's first law of motion is that when you exert a force, things accelerate, and then they go on moving forever in a straight line and they do not deviate or slow down until you exert another force. But I would contend that actually we biologists explain things very much by Aristotelian principles. We have enzymes do things, they push things, they pull things, and that is how you understand them. So in a funny way, we are very far from the underlying philosophical underpinnings of our subject and I am thankful to say that quantum mechanics on the whole does not play a great role in biology.

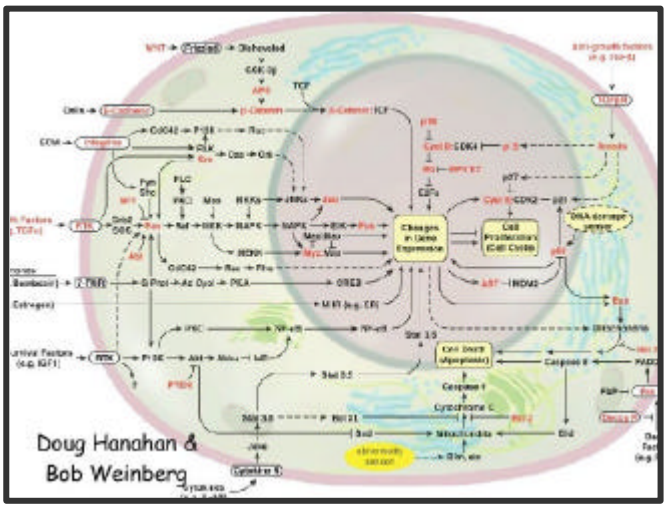
Another example of this same kind of thing is that a niece of mine came to work in the lab and I asked her how many molecules she thought were in this glass of water. She looked completely blank. Then I realized that I had no idea how one knew what the value of Avogadro's number was. So I asked a friend and he said, "oh well, that was Einstein's Ph.D. thesis" – and so indeed it was. This great French physicist, Jean-Baptiste Perrin, won the Nobel Prize in 1927 or 1928, and he did experimental measurements of Einstein's Brownian motion. He wrote this beautiful book called *Les Atomes* (which I am glad to say you can get in an English translation), published in 1913. The book is devoted to proving that atoms really exist and this is actually the denouement of the book, so these are all values of Avogadro's number – the viscosity of gases, Brownian motion, critical opalescence, radioactivity, and so on and so forth. My favorite by far is the blueness of the sky: direct sunlight is five million times brighter than the blue sky, and because of that, Avogadro's number is 6×10^{23}. You have to be a really good physicist to be able to

deduce that. It is really curious that this ancient physics is so inaccessible to modern biologists.

What I am going to talk about is something that is, by comparison, utterly simple, utterly childish and utterly straightforward, and yet extremely complicated – in a funny way, much too complicated for physics to comprehend. As we know, physicists cannot even solve the dynamics of three bodies! Two bodies are okay, but three are not – it tends to show chaotic behavior. What can we say about this?



First of all, everything is made of cells. Those cells have a number of options, and the key to understanding the growth of an egg into an adult organism, or a cancer when it develops, is to understand the transitions between these various states of dividing and not dividing, growing and not growing, or dying if the conditions become too stressful, and the recycling of the materials. It is not a trivial problem as I will show.



We know an awful lot but it is extremely difficult and the challenge for the future is to understand how all these signals are coming in from the outside. It is a little difficult to see on this slide but there is a cell here, its margin is around here, then there is a nucleus and things are going on into the nucleus. There are all these growth factors which molecular biology has successfully solved, but what we really do not understand is how you can understand how the changes in gene expression actually impinge on the cell cycle engine, which I am going to talk about during most of this talk. I think this is a really serious challenge and I am hoping that mathematicians and physicists and bioengineers will make a contribution to making sense of the individual atoms, if you like, the enzymes in this pathway, how these are integrated to explain the shape, the size, the growth or lack thereof, of the cells.

Why do cells divide? Well, the truth is that cells only have a limited capacity for growth and they have limited life spans, and the only way to overcome these limitations is to grow and divide. There are very few cells in our bodies which do not replace themselves in the course of a lifetime. A very interesting question that I do not understand the answer to is why the germ line is apparently immortal whereas our bodies are all too mortal. I once asked a friend who works on this why this was and she said, "This is because of natural selection and evolution," and I

said “yes, but what is the molecular mechanism whereby the germ cells are immortal?” “Aha,” she said, “that is for you to figure out, Tim, you are the molecular biologist.” So, we do not know.

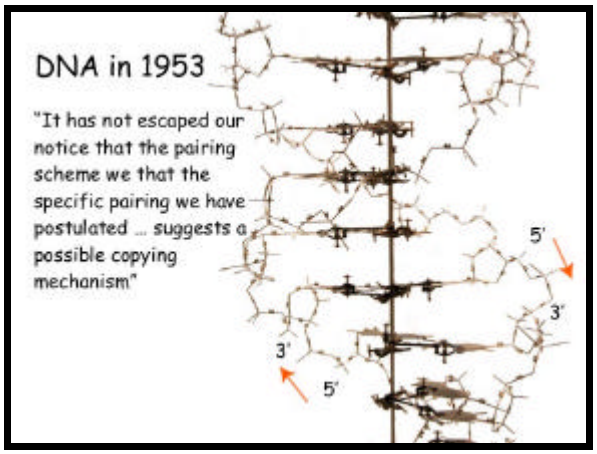
I think the way you find out things in biology is to do experiments. In my view those experiments consist solely of measuring something and doing lots of controls, and the controls must be positive and negative.

In the context of this meeting, I want to show you that things take very much longer; the fog often takes a long time to be dispelled. This is a quotation from the great English physiologist JBS Haldane and he says that forecasting the future – and this is a quite general law to be noted – the unexpected always happens, so one can be sure that the future will make any detailed predictions look rather silly.

Haldane actually was very prescient because in a little magazine published in the biochemistry department in 1931, he wrote down this joke exam question which was what the students of 25 years in the future were going to be solving. The remarkable thing is that I think he was about 10 years out – if he had said 1966 instead of 1956, he would indeed have been able to write down the structural formula of haemoglobin. In 1931, this was a pure pipe dream: enzyme action is only intelligible in terms of wave mechanics – absolutely brilliant. This is exactly what we now do – except rather than the biochemistry of the terpenes, we would say it is the sequence of the DNA. These people in the 1930s were amazingly foresighted about what the programme they were embarking on was going to tell them, but it took a hell of a long time between the ambition and the fulfilment of the ambition. In a way, we are still working that programme out. I think we are in the beginnings of coming to something that is absolutely new, as I tried to say a moment ago.

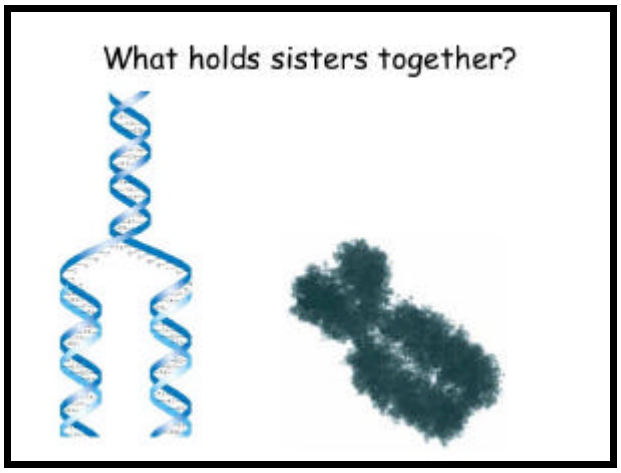
Science is a game, Schrödinger said, but a game with reality. One should really remember that the playful aspects are extremely important. I know that here there is an ambition to generate real things out of science, but I think the best things that come out of science, that basic scientists like me can provide is a sort of spirit of adventure, a spirit of playfulness, because who could have dreamt, for example, that studying the flashing jellyfish would be such a useful tool for cell biologists today? That kind of playfulness I think is completely impossible to see. I was reminded last night of a quotation from Michael Faraday who, when a woman came up to him after one of his famous lectures in the 1830s, and asked, “Mr. Faraday, what could possibly be the use of this electricity you have told us about this evening?” He said, “Madam, of what use is a tiny baby?”

So, Schrödinger’s programme was to ask is it possible to explain life in terms of physics and chemistry? As we know, his little book, *What is Life?,* inspired the early molecular biologists to get away from a vitalist kind of obscurantist explanation of life and actually reduce it to molecular reality.

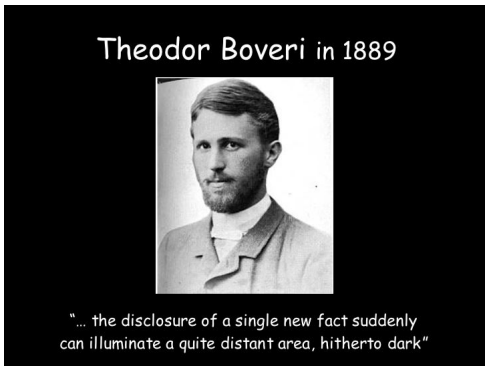


One of the pioneers in that was Jim Watson – next slide, please – and Francis Cricks' discovery of the two-strandedness of DNA which immediately explains why cells divide into two and not into three, as I suppose they would have done if the triple-stranded helix of DNA had actually been correct.

So to understand the basis of cell division, one really has to go right into the centre and talk about DNA replication. The extraordinary thing is that we really do not need to know the details of what happens at the replication fork and they are still being worked out. There is an amazingly complicated nano-machine that takes place here to bring about this synthesis of the two strands.

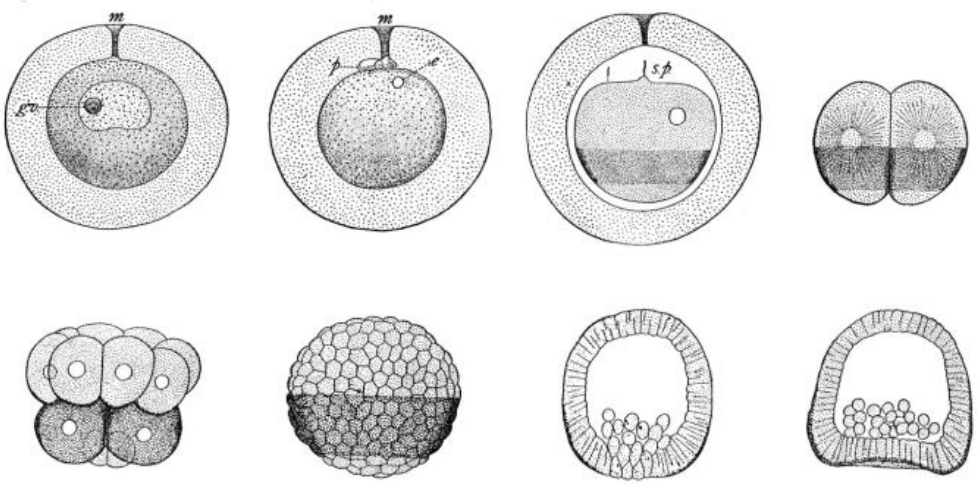


In order for the faithful inheritance of the DNA to be transmitted to the two daughter cells, it is absolutely crucial that the two daughters of duplication be held together, and we now actually understand rather well how they are held together: they are tied up by little strings of protein.



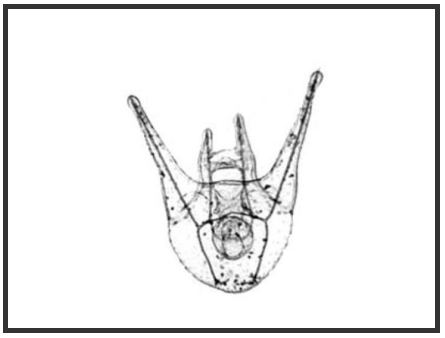
The first person to really understand how this went on was this mannered young German biologist, Theodore Boveri, and he did his great experiment which shed light on it in Woods Hole and Naples in 1902.

Sea urchin development

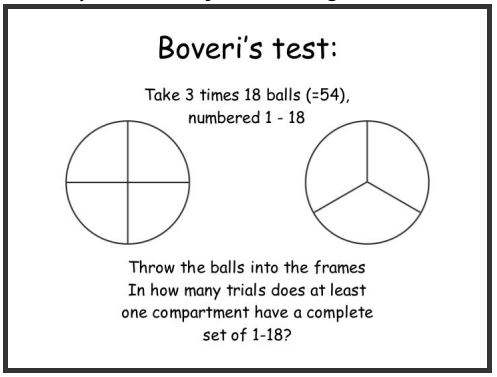


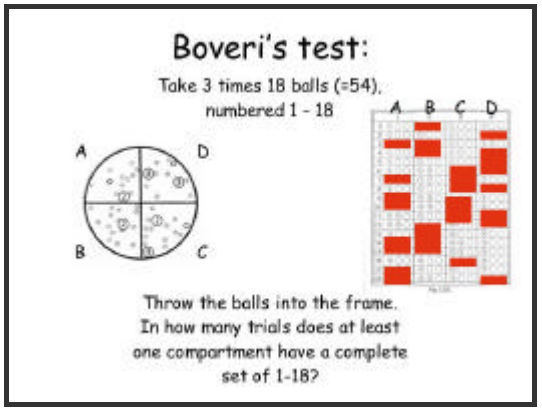
Boveri

He was studying sea urchin eggs. Here is a sea urchin egg being fertilized and then dividing rapidly to form a hollow ball of cells. About three days later it gets a mouth and an anus and is called a pluteus larva. Notice that this and that are exactly the same size – this will actually probably be slightly smaller than that because these creatures do not eat. It is simply a matter of the reorganization of the materials in here that gives rise to this beautiful swimming larva.



Apparently, science was just as competitive back then as it is today because Boveri wrote, I think to Thomas Hunt Morgan, and said, "I have just found out this fantastic thing. It is such an easy experiment, so please do not tell anybody because anybody could be reproducing it and then I would lose my priority." It was not a matter of loss of priority really because somebody had done this about 20 years before but did not have the genius of Boveri to have an insight into what was going on. If you use too high a concentration of sperm to fertilize a sea urchin egg, at the first division it divides into four instead of into two. If you shake the suspension of eggs very hard, then some of those eggs divide simultaneously into three rather than into four. By the way, after this, these cells now divide into two, as usual, and likewise here. What Boveri noticed was that hardly any of these formed a swimming pluteus larva whereas about one in twelve of those ones did. Then he wondered why that was. What he did was to imagine what the role of these chromosomes was. This had already been described and their behaviour during mitosis had been noted, but Boveri wondered whether it was the total amount of DNA or the actual kind of DNA that was important. He counted the chromosomes and, went back home to do a simple experiment. In fact, he asked a theoretical physicist, "What is the probability of a single cell dividing into four getting a complete set of chromosomes?" The physicist said, "Well, the statistics are a little tricky to work out, so why do we not do the experiment?" So they did the experiment, throwing the balls on the ground, putting down these formers and counting how many balls fell in each of these quadrants.





What they found was that when they did that and they counted the chromosomes – here are the four blastomeres, one, two, three, and four – where I put a red dot they did not get a chromosome, so the furthest blastomere is missing chromosome 3, missing chromosome 7, 9, 10, 14, 15, 17 and 18. You can see that not one single cell got a complete set of chromosomes. When they did this – and they were very thorough and Germanic about it and repeated it thousands of times – they found that the statistics matched perfectly Boveri's observations on the percentages. Three genomes into three cells is obviously a much higher fraction of completeness.

So Boveri concludes from these experiments that only one possibility remains: namely that not a definite number, but a definite combination of chromosomes is essential for normal development, and this means that the individual chromosomes must possess different qualities. He then went on to point out that in tumours they

Thus, only one possibility remains, namely that not a definite number, but a definite combination of chromosomes is essential for normal development, and this means that the individual chromosomes must possess different qualities.

very often saw cells that looked like his polyspermic urchins and he muses in this great paper of 1902 that maybe multi-polar mitoses might under certain conditions lead to the development of tumour-like formations. In this, he was absolutely correct – they undoubtedly do. The curious thing is that although we know so much about cell division, we still do not know very much about the duplication of Bovari's favourite things, the centrosomes, the spindle pole bodies, and we still cannot account for this curious behaviour of tumour cells by any rational mechanisms. We do not know whether it is because of a failure to complete cytokenesis or a failure to regulate properly the division of those poles.

"Multipolar mitosis might under certain conditions lead to the development of tumor-like formations..." Boveri, 1902

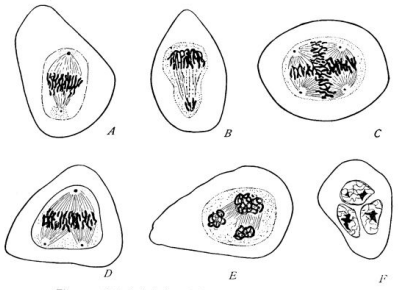


Fig. 32. — Pathological mitoses in human cancer-cells. [GALEOTTI.]
A. Asymmetrical mitosis with unequal centrosomes. *B.* Later stage, showing unequal distribution of the chromosomes. *C.* Quadripolar mitosis. *D.* Tripolar mitosis. *E.* Later stage. *F.* Tri-nucleate cell resulting.

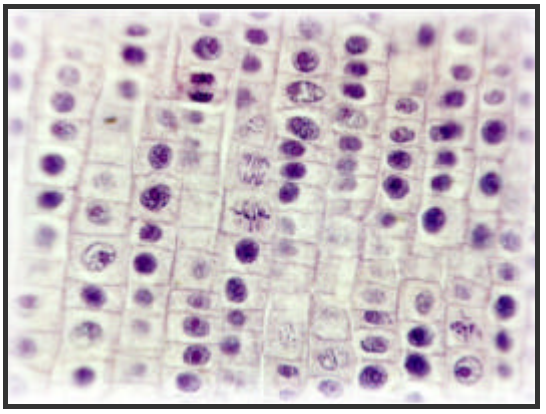
Of course, there is another way that you can get bad chromosomes. It is very important to emphasize that both of these discoveries were a very long time ago. This one is about 1931. You look at these chromosomes from a tumour cell.

You now paint them by modern technology and you can see that they are completely scrambled. Almost every chromosome is made up of two chromosomes by a process that was discovered by Barbara McClintock studying maize I think in Indiana or Iowa or somewhere like that. Not much to do with human disease, but she understood it perfectly.

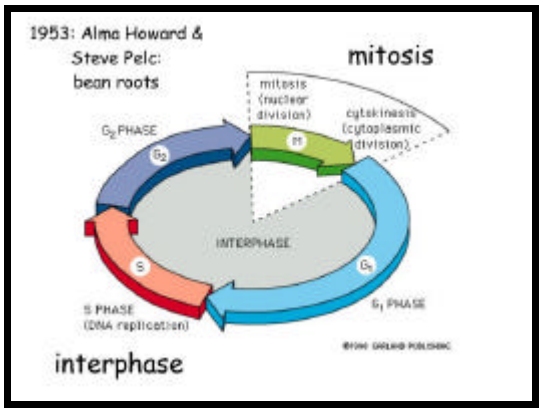
If chromosomes are irradiated, they tend to break. When they break, they join together, and when they join together they can be pulled apart because of having two centrosomes. Her paper on the breakage-fusion-bridge cycle was tremendously prescient and informative about the causes of human cancer – not from studying human cells, not from studying cancer, but just from studying the normal inheritance of chromosomes in maize.

By 1944, almost 50 years after Boveri had discovered the importance of the chromosomes, the basic mechanisms of mitosis were very obscure. People worried about things like the forces that moved the chromosomes and discussed whether the mitotic spindle that they had seen down the microscope was real or an artefact. I love this rather pessimistic and chest-beating account of Franz Schrader

who worked at Columbia in 1944 and wrote this nice book on mitosis: we failed so badly, we just do not understand what is going on.

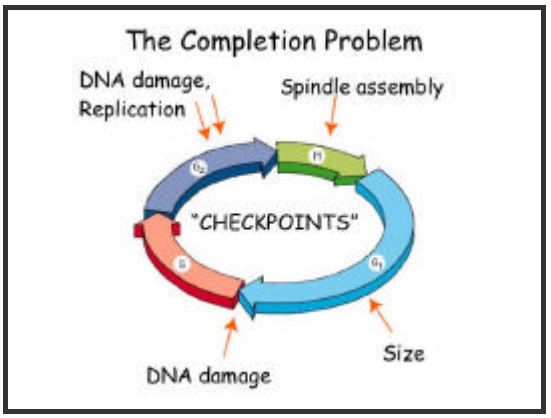


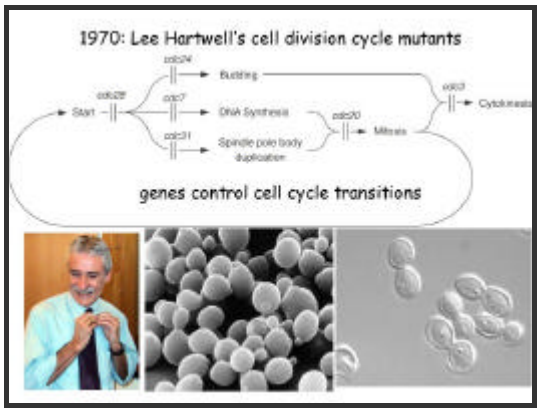
The real advance came when radioisotopes became available and people began to worry about the effects of ionizing radiation on cells. Now we move into the clinic, because the key experiments here, the classis ones, were done by two workers, Elma Howard and Steve Pelch, who worked in a radiation clinic but were encouraged by their enlightened boss to work on broad beans. This is a picture of a broad bean root and you can see that the chromosomes condense in a very high fraction– in fact all of these cells are involved in dividing. Howard and Pelch discovered that the bean roots took up radioactively labelled phosphorous and that they could label the DNA thereby.



Namely, that DNA synthesis took place at a discrete interval in the cell cycle, completely out of phase with mitosis – there were long gaps between the end of the previous division and the initiation of DNA replication in these cells.

They also discovered the concept that we now call checkpoints. They found that when they shone X-rays at the cells that were in this phase of the cell cycle, they failed to start DNA replication. If the X-rays, on the other hand, were shone at the cells later on in the cycle, they failed to enter mitosis. You will find if you go back and look at their 1953 paper that they had a very clear appreciation of this point. Of course, they knew nothing about the underlying molecular biology.



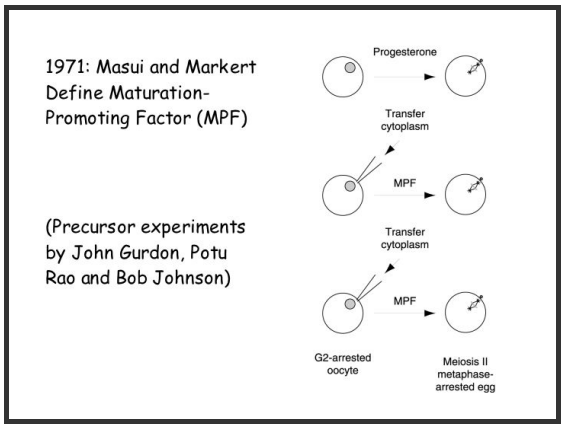


Their problem, however, did raise an acute question as to why: whether this was some kind of hard-wired business or whether this was achieved by biological controls, so to speak, in software. The idea that this is not something intrinsic, even people like Hartwell wondered whether the programme of mitosis required replicated chromosomes to act as a sort of substrate and then the process of mitosis generated individual chromotids, which then had to be replicated.

A moment's reflection, if you are a biologist, shows that there are actually many exceptions to this rule. There is meiosis, where you have two N-phases with no S-phase, or the platelet precursor mega-carrier cells which undergo multiple S-phases with no intervening N-phase. You will be able to think of other examples, where these rules are violated. So clearly, there is some underlying control process to the control of mitosis and the control of DNA replication.

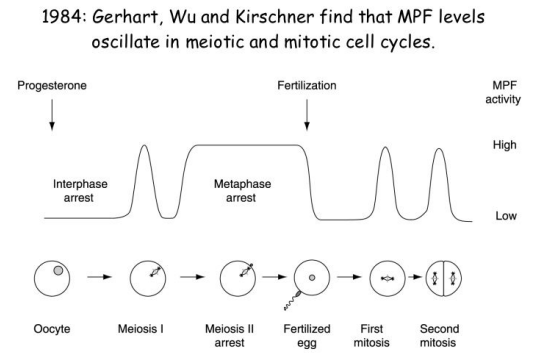
The first important experiments are due to Bob Johnson and Potu Rao, who simply fused cells together at different phases of the cell cycle and made the astounding discovery that any cell nucleus fused with a mitotic cell would enter into mitosis. We will not worry about the other details here because this is the important thing they discovered. Now, interestingly enough, although these experiments showed, in a way, the logic of the cell cycle, they unfortunately did not provide any experimental means of further analysing the molecular basis for this. For that, one had to turn to a different system.

That is the study of oocytes, where Yoshio Masui and Dennis Smith were studying the effect of reproductive hormones on these cells – just the formation of this little white dot. When they fell over, by the way, that is the first meiotic division.



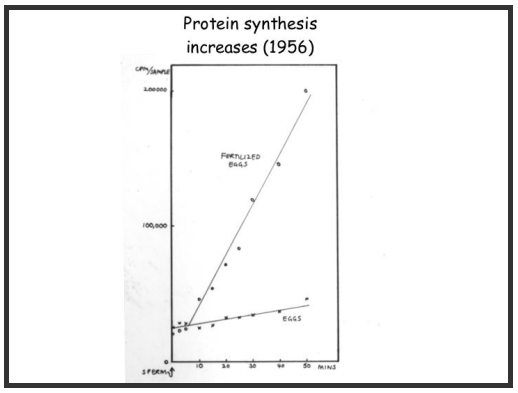
What Masui and Markert discovered was that there was a factor which accumulated in response to progesterone, a conversion from an inactive precursor which did not require protein synthesis, such that you could now suck out a little bit of cytoplasm and inject it into a new oocyte, and those oocytes would recapitulate the process of maturation without the need for progesterone. You could repeat this many times so that there was no possibility that any of the original progesterone molecules survived. They discovered that the transferable factor was a protein, which was heat-labile, protease-sensitive. They and others tried to purify it, without, it must be said, initially very much success.

It was not for another decade or so that John Gerhart, Mike Wu, and Mark Kirschner discovered that MPF was not something just to do with frog oocytes. First of all, it had been discovered by a Japanese group, that starfish oocytes contained this stuff. Then it turned out that dividing human cells contained MPF. The rule always was that whenever MPF was high, the cells were in the process of division, whether meiotic or mitotic, it did not matter. MPF oscillated with the cell cycle, up and down. When I heard about this from John Gerhart, who came to give a talk at Woods Hole in 1979, I was fascinated by the idea that there could be an enzyme that catalyzed mitosis and I wished that I was working on it, but in fact I was not working on anything remotely like that at the time.

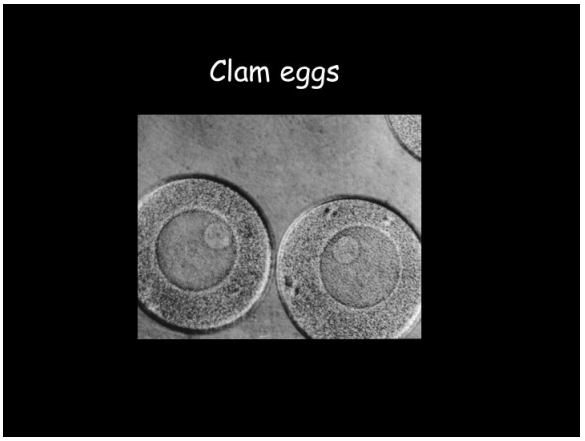


Getting sea urchin eggs

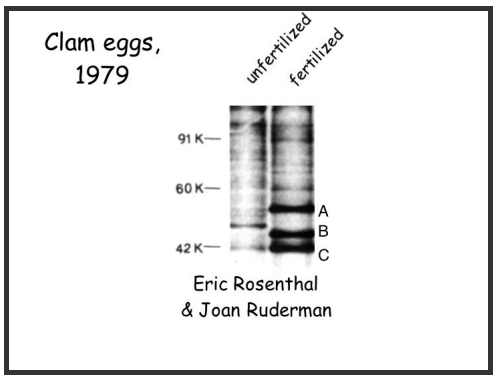
In fact, I went to Woods Hole to study protein synthesis in sea urchin eggs. These sea urchins are very measly sea urchins – they are right at the northern end of their range and they are not very healthy. It is a shame that this is a very old photograph and it is not a colour photograph, because you would see that the eggs of this particular species, Arbacia punctulata, are a beautiful deep red colour. You get the eggs out by giving them a 12 volt AC electric shock. We do not know if the urchin feels pain or not, but probably not because it does not have much of a nervous system, in fact.



This is what I wanted to understand. When you fertilize the eggs, there is a huge increase in the rate of protein synthesis. I struggled to understand that and worked on it for about five years in Woods Hole, actually without making any serious progress at all. Luckily for me, in 1979 as well, I encountered the following organism.



The surf clam; spisula solidissima.



This was being worked by two friends, Eric Rosenthal, a graduate student, with Joan Ruderman. We collaborated a little bit because what they found was that when clam eggs were fertilized, there was not much of an increase in the rate of protein synthesis, but they started making new proteins. These three bands here are clearly absent or very much reduced in the unfertilized eggs. It turned out that this was an example of translational control, which at the time was not clearly something that could happen. Again, we were very interested in the mechanism of translational control.

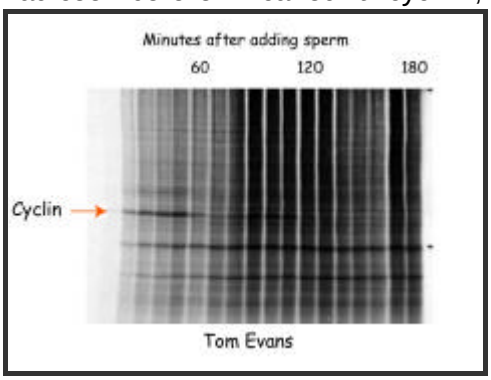
More proteins (urchins),
or new proteins (clams)…

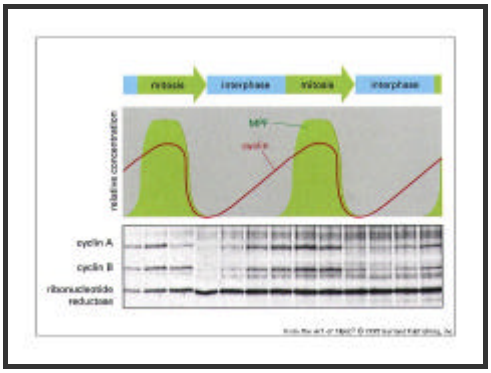
Inhibiting protein synthesis
blocks cell division…

We began to wonder why did sea urchin eggs make more proteins and why did clams make new proteins after fertilization? I became aware that people had long known that if you inhibited protein synthesis that even the very first cell division did not take place. Although most of the experiments we did involving mashing things up and adding detergents or adding acid and precipitating and counting them, there were rather good microscopics around, so I started looking at cells.

In 1982 I did an unbelievably simple experiment which was designed to do something completely different and the protocol was very simple: check if the eggs are fertilized, that is, the envelopes, then add the radioactive amino acids, which are precious; sample at regular intervals, because that was what one did in those days; dissolve the samples in sample buffer and analyze on a gel.

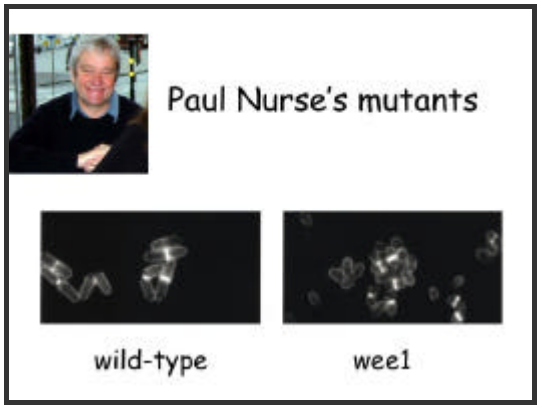
What I saw, to my astonishment, was the behaviour of this protein which nobody had seen before. I called it 'cyclin', because it went up and down in time with the cell cycle. It was one of the first proteins that you could clearly see as the discrete band on a gel – it went up and then it just faded away. Then it came back again. We could show that it kept on coming and going as long as the cells went on dividing. We could show that you could stop it coming and going by inhibiting cell division or inhibiting DNA replication. So there was clearly a connection between the behaviour of this protein and the cell division.



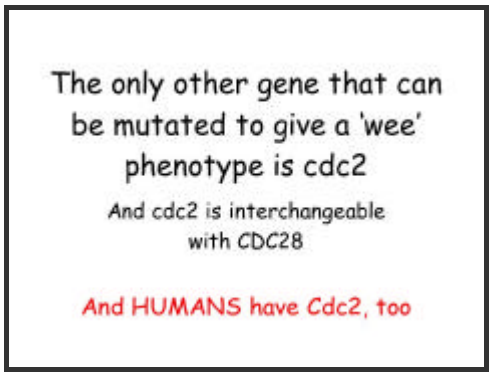


It looked much as though it somehow described MPF. When we looked in clams later on that summer to see how universal it was, we were astonished to find that these proteins, which we had thought of as being simply interesting translationally regulated ones, also went up and down in the same way, slightly out of phase. You did not need to be a genius to anticipate that there might be some connection, although we worried a lot about how a continuous increase here could be translated into the rather abrupt transition that entering mitosis, and worrying about that actually set us back a little bit.

So we sent a paper in, to sell, and we got this wonderful referee's comment, "This is wild speculation based on faulty logic." The editor said, "Dear Tim, thank you for sending in your paper. The good news is that we will accept it, but the bad news is, in nothing like its present form." So it was rewritten and accepted. Remarkably, people did not know what to make of it and people thought that it was impossible to suggest this almost heretical possibility that such a thing as a protein coming and going. It was especially the 'going' that was incomprehensible to everybody at the time. Remember, the Ubiquitin system had only just been discovered and was seen in terms of a way of getting rid of abnormally folded proteins from cells. The idea that it could be highly specific and selectively degrade proteins was still almost a decade away from being discovered.



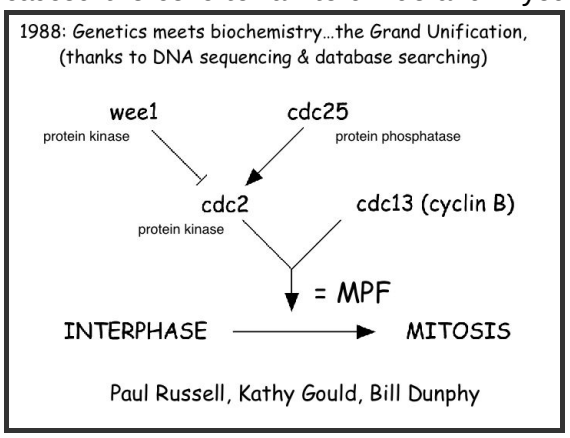
I made what I realized at the time what I thought was undoubtedly the greatest discovery of my life. Remember, I showed earlier a quotation which said that really important discoveries are important precisely because of their unexpectedness. You cannot contract to make an important discovery; all you can do is hope. So I changed fields completely – obviously not overnight – and I began to find out about what was known about the control of the cell cycle. The interesting thing was that quite a lot was known, thanks largely to the work of Lee Hartwell, a great pioneer, and Paul Nurse. Lee discovered that there were genes that controlled the cells. He also discovered this completely by chance – by looking at the yeast cells down the microscope he discovered that certain mutations gave rise to a very uniform phenotype because an actual process to do with cell division could not be completed. He accurately diagnosed that this particular gene here, CDC-28, was a kind of a master regulator because it controlled three quite different biological processes simultaneously. But one did not know how to analyze that further because in 1970 the possibility of isolating genes and sequencing them was five or six years away.

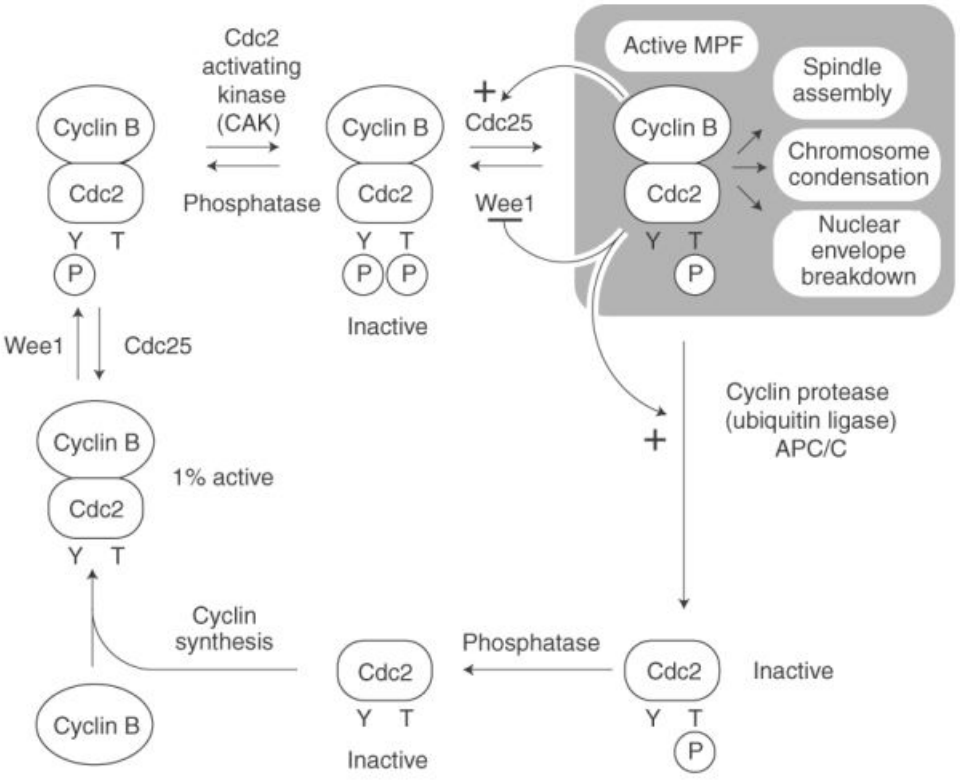


Paul Nurse also wanted to study cell division and wisely chose a different yeast – a yeast in which it is easier to measure the size just by measuring the length. He was delighted to discover, again completely by accident, yeast that divided to smaller than usual size. These are the so-called 'wee' mutations, because 'wee' is Scottish for 'small'.

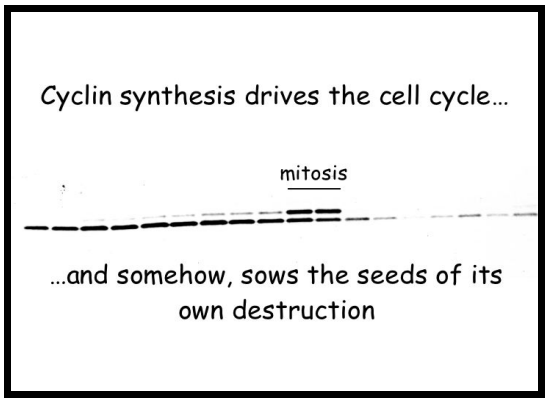
He was so intrigued by that that he went on to isolate more of these kinds of

mutations. Almost all of them mapped in the wee-1 gene, but then he finally found the wee-2 gene, the fiftieth strain that he identified, and this was mapped in a gene he already knew, called CDC-2. This was a gene that if you immobilized, it caused the cells to fail to divide and if you somehow hyper activated, it caused the cells to divide to smaller than usual size. It was discovered then, I think by David Beach, that CDC-2 and CDC-28 were interchangeable, and by this time cloning and sequencing became possible and it turned out that CDC-2 was a protein kinase – although Nurse failed to find any protein kinase associated with it because he used the wrong substrate. The real breakthrough came when Melanie Lee discovered that humans have CDC-2 as well.





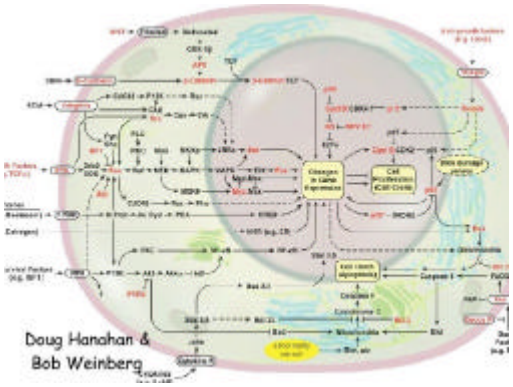
Fred Lohka and Jim Maller finally succeeded in purifying frog MPF and they discovered that it had two sub-units. When I saw this picture, I think in the summer of 1987, I said, "as soon as we have antibodies against frog cyclin, I bet we will find that this band here is cyclin", and indeed it proved so. It was really Marcel Doré working in Montpelier and purifying starfish MPF who showed definitively that this band was cyclin and this band was CDC-2.



The nature of the cell cycle engine, the nature of the enzyme that catalyzed mitosis, suddenly became quite clear. CDC-2 combines with the cyclin and that catalyzes the transition from interphase into mitosis.

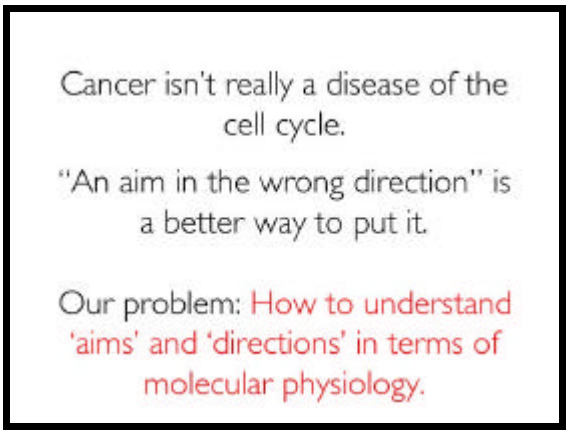
That is what it looked like. Now I will discuss the quantum mechanical explanation of this.

Now, it is really complicated because there is a residue here that has to become phosphorylated and as the result of that phosphorylation, in comes the substrate, the ATP bends down to meet it, the serine attacks that phosphate and off it goes. This is how it is seen. A huge number of people are involved getting this straight. The truth of the matter is that we now understand this cycle rather well, but there are two aspects of it which are still deeply mysterious. How is it possible for one enzyme, one protein kinase, and actually there are a couple, aurora and polo, which seem to collaborate in important ways with CDC-2 in higher organisms at least to catalyze all the changes in time and space that comprise mitosis. We still do not understand this very well and we still do not understand the control of proteolysis very well. I wish I had more time to tell you about what we do and do not know, but it is still an incomplete story.



The future challenges for the field are not so much about the nature of the cell cycle regulator, but how cell growth is regulated. You notice that, apart from the first slide that says cells can grow and divide, I have not said anything about the control of cell growth because surprisingly, this is still very mysterious. If you go to a growth factor meeting, it is just beginning to unfold. How growth factors actually make cells grow is something that is not well understood. It basically boils down to the control of ribosome biosynthesis. What determines the cell size is something completely mysterious and more important is the problem of cellular homeostasis. I like to say this is a problem of your nose: you are born with a nose and it grows to an adult size, and the amazing thing is that your nose just stops at a certain size – it neither shrinks back into your face, neither does it grow out. How does a nose know how big it should be? Cells are dying and dividing in there all the time and the nose stays the same size. It is a really profound problem in biology.

One question you could ask about this in the context of a biotech associated organization is "will CDK inhibitors cure cancer?" Obviously if you could inhibit these enzymes, you could stop the cells dividing. The trouble is, of course, if you stop the cells dividing, this is not a very good thing. If you stand too close to an atom bomb when it explodes, your cells will stop dividing and you will die in about a week. So the question is where the specificity of such inhibitors might come from. Some people say that cancer is a disease of the cell cycle, but I would say it is more a disease of cell differentiation and cell growth rather than the cell cycle. If cancer was really a disease of the cell cycle, then there would not be any cancer because the cancer cells would not be able to divide, and unfortunately, they can – they divide all too well.



I think the problem of cancer is really a problem for developmental biologists rather than cell cycle people. It is in going back to understanding these transitions between the cells and how the growth factors do it that the future lies – to my way of thinking, understanding growth factor inhibitors. Indeed, there is one real success story in the biotech industry. The monoclonal antibody that reacts with the erb-b2 receptor, herceptin, is a very good success story, where if cancer cells have a very high level of that receptor, monoclonal antibodies actually stop those cells dividing. This really helps women with these particular, admittedly rather small, class of tumours.

This is the challenge that meets us. To understand not the molecular nature of these pathways – that is really quite understood – but to understand the coordination of all these things: how the cell integrates the information to changes in gene expression, to changes in cell growth and to changes in cell proliferation. I do not expect the advances to come either predictably or necessarily soon, although one never knows, but the sure fire thing is that as long as people keep working on this, steady progress will be made. I think there is a great future for young biologists today and I think there is also a great future for young biologists to get together with their mathematical and physicist friends to try and work out the new paradigm to make sense of the complexities that we know as the cell.

Thank you very much.

Question (in French) : Do you have a recipe to make biologists, physicists and mathematicians work together ?

Tim Hunt
The question is how to get these people together. The answer is that people are beginning to get together. My friend Andrew Murray, who is a great pal and worked on the cell cycle, is at Harvard University trying to promote such exchanges. Again, another person I know at Harvard, Mark Kirschner, is changing

from being a department of cell biology to becoming a department of systems biology. Mark's friends tease him about this because actually systems biology really means old-fashioned physiology, but they are very actively looking for physicists. There are one or two people who are interested in these problems. There was in my youth, and there probably still is, a journal called the Journal of Theoretical Biology. I never found anything very useful in the Journal of Theoretical Biology, but I think things are changing. I think the important thing is that it is absolutely critical that the theoretical biologists do experiments as well to confirm or refute their hypotheses.