A modern day gene genie

Sir Richard Roberts, Nobel Prize winner and founder of REBASE - Restriction Enzyme dataBASE - gives us a snapshot of his work with restriction enzymes

Can you provide an overview of the aims and focus of the research activities of REBASE?

The main aim of REBASE is to maintain a comprehensive listing of all genes and proteins associated with restriction-modification systems. Originally, this was restricted to literature and personal descriptions of new enzymes, but as DNA sequencing became important, the genes encoding known restriction system components were included. Later, putative restriction system components could be identified by bioinformatics, so these too are now an integral part of the database.

How did you first come to be involved in research around restriction enzymes?

In early 1972 I heard a lecture by Dan Nathans at Harvard Medical School in which he described the use of the first Type II restriction enzyme, Endonuclease R, to cleave SV40 DNA. This seemed to me to hold the key to sequencing DNA and when I moved to Cold Spring Harbor Laboratory later that year I immediately began purifying this enzyme and the few other restriction enzymes that had been discovered in the interim. The connection to NEB came later.

Roughly how many restriction enzymes are known at this time? Do you expect this number to keep growing?

More than 300 unique specificities have been characterised, as well as several hundred isoschizomers (enzymes that recognise the same sequences as some of those), but by examining the DNA of the sequenced bacterial genomes we know that many, many more exist. It is likely that many thousands, perhaps hundreds of thousands, are present in nature.

How would you describe the ethos of New England Biolabs? In the 35 years since its inception in the mid-1970s, would you say the ethos has changed?

The original vision of Don Comb, the founder of NEB, was to make money to support research. I shared this vision completely and so it was a perfect synergy for me to work with him to make restriction enzymes the first major commercial product of NEB. This is still the overarching vision of the company. The fact that Don is 83 and still an active research scientist, together with the many other researchers who contribute to the success of the company, is what has allowed us to maintain that original vision. Of course the fact that we are still a private company and not subject to the whims of shareholders, who typically feel that research should stop to increase their profits, has been a major help.

Relating to your Nobel Prize in 1993, for your discovery with Phillip Sharp of 'split genes': could you describe why it was so significant?

The significance stems from the fact that it completely changed our view of what constituted a gene in eukaryotes (higher organisms). It had repercussions throughout eukaryotic biology and was of crucial importance for interpreting the human genome sequence, for example. It meant that all higher organisms had an extra degree of complexity in the way in which they used the information encoded in their DNA. Even today we are

Splitting the building blocks of life

Since their discovery, restriction enzymes have proved key to unlocking the doors of modern genomics and molecular biology. **REBASE** is a modern tool to help scientists keep track of these wondrous proteins, while New England Biolabs is an ethical company that supplies them to researchers

SINCE DNA WAS first discovered by Crick and Watson in 1953, massive leaps have occurred in the fields of genetics and molecular biology. One such leap was in 1970, when Type II restriction enzymes were first isolated – a breakthrough that opened new doors into our understanding of how DNA is manipulated in nature, and led the way for DNA sequencing and mapping, and later recombinant DNA technology – techniques which have since transformed modern medicine.

Type II restriction enzymes, or restriction endonucleases, are found in bacteria, and have the ability to recognise sequences in DNA, cutting these with exquisite precision to produce specific fragments (as opposed to Type I restriction enzymes, which cut DNA randomly). In nature, bacteria use these as a defence against invading viruses (bacteriophages), whereby these enzymes cleave foreign DNA in order to prevent invasion, while simultaneously a methylase enzyme adds a methyl group to the restriction site of its own DNA, thus preventing it being attacked by its own defences. This system is known as restriction-modification. In 1971, Danna and Nathans were the first to realise that the small fragments produced by restriction enzymes could be used to map DNA molecules and hence the cleavage sites could be used as landmarks with which to track genetic traits.

THE ROOTS OF REBASE

One man has taken the work of Danna and Nathans and pushed it to new heights. Sir Richard Roberts, the winner of the 1993 Nobel Prize in Physiology or Medicine (with Phillip Sharp) for his groundbreaking discovery of split genes. Inspired by a lecture given by Nathans himself in 1972, Roberts was also instrumental in isolating most of the world's first known restriction enzymes. As a pioneer in this field, requests for lists of these enzymes came flooding in, eventually leading to the formation of a database named REBASE (Restriction Enzyme dataBASE), containing a listing of not only all the restriction-modification enzyme systems, but also the genes encoding these.

REBASE has since evolved into a web-based format (rebase.neb.com) with a searchable interface and a plethora of invaluable information. New developments have meant that categories are added to the database as appropriate. An example of this occurred when the Haemophilus influenza genome was sequenced in 1995, leading to the inclusion of putative restriction system components. Technological advances have led to users being able to BLAST (use a Basic Local Alignment Search Tool) new genome sequences to find restriction system components, and

THE REBASE WEBSITE HOMEPAGE

still learning more about how split genes are processed and finding additional complexities in the regulation of the information present in these split genes.

What did winning the Nobel Prize mean to you as a scientist? Is it something you aspired to or had imagined ever being awarded?

I think every scientist dreams that perhaps one day they might make a big discovery, but few aspire to win a Nobel Prize. There are very many discoveries being made all the time and it is difficult to know when any one discovery becomes more important than another. Luck plays a very significant role. As was famously said by Isaac Newton: "If I see further than others it is because I stand on the shoulders of giants". This aptly summarises all research.

Clearly dissemination is at the core of REBASE – how important do you consider disseminating and sharing of research for scientific progress?

Dissemination and sharing are key to all research, but especially so when databases have been compiled and the database managers want as many people as possible to see the results of their work. Much of the dissemination of the contents of REBASE has been in response to user requests over the many years that I have been running it.

INTELLIGENCE

REBASE : THE RESTRICTION ENZYME DATABASE

OBJECTIVES

This project concerns the maintenance and distribution of a cohesive and comprehensive database of information about restriction endonucleases and their associated methyltransferases. Key components of the database are information about the methyl sensitivity of restriction endonucleases and a comprehensive analysis of the restriction enzymes present in the sequenced microbial genomes. REBASE is a unique resource for the practice of biotechnology and its contents are used directly or indirectly by many sections of the research healthcare community. REBASE serves as the master database used by both academics and commercial companies to find out which restriction enzymes are commercially available.

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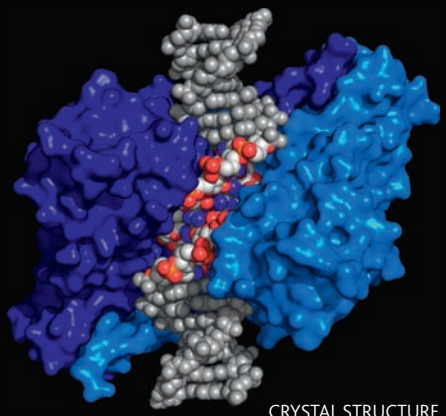
DR ROBERTS was educated in chemistry at Sheffield University and molecular biology at Harvard University. He worked for 20 years at Cold Spring Harbor Laboratory where his group discovered RNA splicing for which he was awarded the Nobel Prize in 1993. In 1992 he moved to New England Biolabs as Director of eukaryotic research before becoming Chief Scientific officer in 2005. He has had a long-standing interest in bioinformatics, which most recently had been applied to his research on restriction enzymes.

also help to determine recognition sequences of new restriction enzymes. Other tools include NEBcutter, which produces lists of user-submitted DNA sequences and maps of cutting sites. In addition, monthly lists of known enzymes and details of their commercial availability are sent out to researchers and interested parties. The formation of such an interactive and comprehensive database requires a variety of means: "REBASE depends on both biochemical and bioinformatic methods to produce its contents," Roberts explains.

NEW ENGLAND BIOLABS

While Roberts is a pioneer in his field, and also at the forefront in the dissemination of everything currently known about restriction enzymes, he also plays a major role in the commercial side of his and others' discoveries. He is currently Chief Scientific Officer of New England Biolabs (NEB), a company founded by Don Comb, whom he first met in the 1970s. At that time, Comb had set up a small laboratory with his wife and a technician, and was planning to start selling the first restriction enzyme. With a large collection of his own, Roberts soon joined forces with Comb as their chief consultant. The rest, as they say, is history, as NEB was the first company to sell restriction enzymes and other research reagents that have since driven the biotechnology industry.

As the leader in high quality research products, and the largest supplier of restriction enzymes in the world, one might expect NEB to be run on traditional, profit-driven corporate values. However, even in such a fiercely competitive and often cut-throat world, the company has managed to remain not only successful, but has been able to do so without compromising their original principles: to create revenue that helps fund research. It is still a private company that believes in sustainable, ecological practices. Comb remains actively involved in research, but has handed over the day-to-day running of the company to James Ellard. Roberts is well aware of the road ahead for such an ethical operation: "The challenge for the future will be to maintain the company ethos and make sure that we continue to provide top quality reagents for research," he muses.

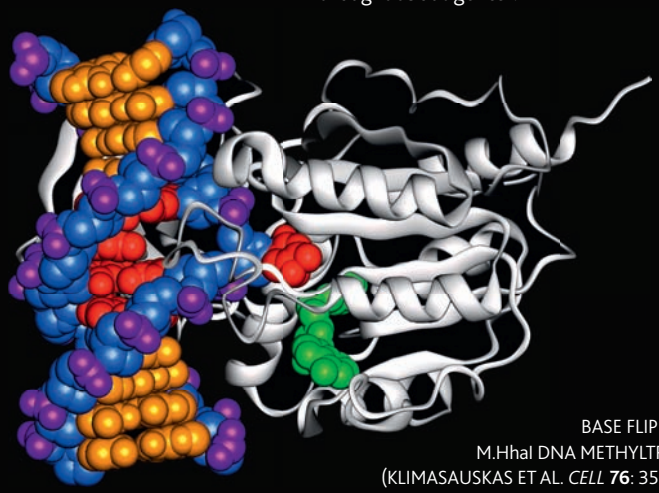


CRYSTAL STRUCTURE OF NotI BOUND TO ITS RECOGNITION SITE. LAMBERT ET AL. *STRUCTURE*, **16**: 558-569, 2008

The ethical work of NEB is also mirrored in the NEB Foundation, which was set up by Comb in 1982. It is an independent, private venture which supports grass roots organisations that encourage conservation of biological and cultural diversity, maintain terrestrial and marine ecosystems, and support local communities, as well as promoting arts projects.

INSPIRATION

As an eminent scientist and Knight of the Realm, Roberts is quick to acknowledge those who inspired him to follow the path that he ended up taking. From his headmaster at primary school, Mr Broakes, who engendered his love of mathematics, to Professor David Ollis, who introduced him to the joys of organic chemistry by challenging his then potentially brilliant young mind. He is also thankful to author John Kendrew for bringing to life the world of molecular biology after reading *The Thread of Life*, while his post-doctoral advisor, Jack Strominger, mentored and encouraged him to follow his dreams. Furthermore, he is grateful to Uttam RajBhandary at MIT for teaching him about tRNAs and research techniques. Ever humble, he puts across his views on what would be the pinnacle of any scientist's career – the Nobel Prize: "Phil and I were lucky to be working in the right area, at the right time, and to make the critical observations that led to what turned out to be a very important discovery because it completely changed the way biologists thought about genes".



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