

Yeast genetics and genomics

Alan Wheals

Research into yeast genetics has brought great insights into eukaryotic cell biology. Alan Wheals describes how yeast genomics is opening up even more exciting possibilities in microbiology.

There are two types of geneticists – those who attempt to understand the nature of inheritance and those who use genetics as a tool to understand biological problems. Yeasts are important in both of these areas of research. The genetics of baker's yeast, *Saccharomyces cerevisiae*, only took off in the second half of the 20th century, but it quickly proved essential for studying fundamental aspects of the inheritance of mitochondria and in the analysis of genetic recombination. In the early 1970s, many microbial geneticists turned their attention from prokaryotes to eukaryotes and looked for a suitable 'model' system. *Sac. cerevisiae* was already an obvious candidate and was described as the *Escherichia coli* of eukaryote genetics. The genetic system of the fission yeast, *Schizosaccharomyces pombe*, was also developed and it has complementary virtues. The genetics of both yeasts are now being supplemented by genomics, a new branch of science dealing with the entire genome of an organism.

has been successfully dissecting the complex association of proteins required to 'splice' RNA molecules before they leave the nucleus for translation. Hugh Pelham (Cambridge) has defined the signals on yeast proteins that target them to the right cellular compartments. Yeasts contain prion proteins, analogous to those found in BSE, and are being analysed by Mick Tuite (Kent). This cell biological knowledge is being used by pharmaceutical companies such as Glaxo-Wellcome and Zeneca to help discover and understand new drugs.

● *Saccharomyces cerevisiae* genomics

The most exciting new development is in the science of genomics. Steve Oliver (formerly UMIST and now Manchester) led a European consortium that sequenced the first eukaryotic chromosome (of *Sac. cerevisiae*) in 1992. This led to the creation of an even bigger multinational consortium, under the leadership of the Belgian scientist André Goffeau, which succeeded in decoding the entire genome sequence by April 1996 – the first eukaryote to be analysed. Having the sequence is one thing – understanding it is another. With reasonable confidence, it is possible to identify putative genes within the genome, so called open reading frames (ORFs). There are approximately 6,200 in *Sac. cerevisiae* of which the function of one-third could be described from either previous knowledge or because of a high degree of homology to genes of known function. Another third could not be unambiguously assigned but had features that at least gave some clues to their function. The most surprising discovery was that one-third of the genes were of totally unknown function. Since they belong to no known family, they are often called ORFan (orphan) genes. There is now a worldwide effort to understand the function of all the genes in *Sac. cerevisiae*. The European Functional Analysis Network (EUROFAN) project, headed once again by Steve Oliver, has systematically knocked out the function of approximately 850 genes one by one. Surprisingly, only one-sixth of the knockouts were lethal to the cell. The remainder are being analysed for a very wide range of phenotypes varying from recombination efficiency to the structure of the cell wall. Most knockouts do have a phenotype but often it does not give clear guidance to the underlying function of the gene – this will require further detailed analysis. However, the approach has proved sufficiently successful to have spawned the Yeast Deletion Project in which a European and N. American consortium will knock out all *Sac. cerevisiae* genes for a similar kind of analysis.

Other approaches, particularly in the USA, are being used to gain clues to the function of the genes. Transcriptome analysis has been designed to look simultaneously at the transcripts of 'all' the genes. A synthetic copy of each of the genes is spotted onto a slide in a high-density oligonucleotide array (a DNA

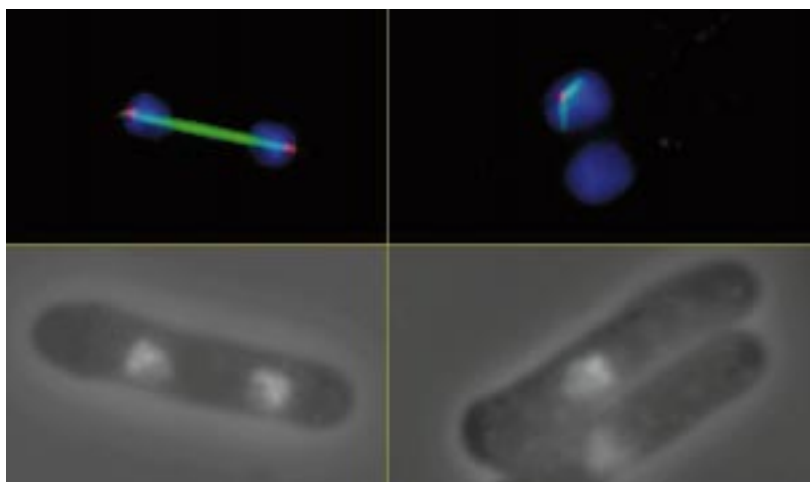
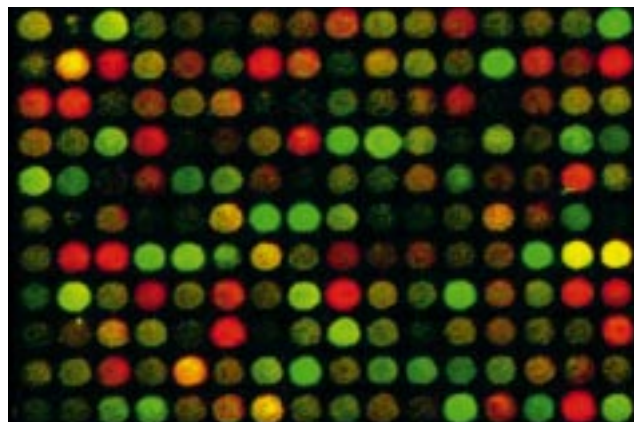


Fig. 1. Mitotic mutant of *Schizosaccharomyces pombe*. The cell outline and chromatin are shown (lower panels) for wild-type (left) and *cut7* mutant cells (right). Upper panel shows microtubules in green, spindle poles in red and chromatin in blue.

COURTESY DOUG DRUMMOND, MARIE CURIE INSTITUTE, OXTEJ, AND IAIN HAGAN, UNIVERSITY OF MANCHESTER

● Significant findings in yeast genetics

What was described by one over-enthusiastic researcher as 'the awesome power of yeast genetics' has, in the hands of experts, provided many of the most important results in eukaryotic cell biology over the last 25 years. There is a very large community of yeast geneticists worldwide but the results can be illustrated with examples of work from some of more than 40 laboratories in the UK. Paul Nurse (ICRF, London) developed the fundamental concept of a universal mitotic control system and the importance of cyclin-dependent kinases. John Diffley (Clare Hall) and Lee Johnston (NIMR, London) have helped define the regulation of the initiation of DNA replication. 'Checkpoints' are required to assess whether a cell can safely proceed through the cell cycle and the molecular basis of this is being determined by Tony Carr (Sussex). Iain Hagan (Manchester) and Robin Allshire (Edinburgh) are analysing genes involved in the mechanism of mitosis (Fig. 1). Jean Beggs (Edinburgh)



chip or micro-array). Fluorescently labelled RNA taken from the cell under a particular set of conditions is hybridized to the DNA dots and the degree of fluorescence gives a measure of the amount of that RNA (Fig. 2). It is thus possible to find out which genes get turned on and off under the conditions of study. These changes in transcription do not reveal what the gene might be doing, but it at least focuses the attention of scientists on potentially important genes.

Some researchers are trying to see the amount and number of proteins present under different circumstances using two-dimensional electrophoresis coupled to mass spectrometry (proteome analysis). Others are looking at how every protein interacts inside the cell with every other protein – a task that, in principle, requires the study of something close to 18 million pairwise combinations. Resources to do these kinds of experiments (Cogeme) have just been established in the UK in Manchester and Aberdeen. This is the era of ‘big science’ in biology – not in the traditional sense of a big piece of apparatus but in the use of robots and automated machines to physically handle the materials and informatics to handle the analysis. However, it will ultimately require the laboratory expert to perform a crucial ‘wet’ experiment to confirm the speculation. It is a sobering thought that it is proving extremely hard even to determine the function of a modest number of genes in a convenient model organism. It will be orders of magnitude more difficult for the human genome project although comparisons between human and yeast genes are already proving informative.

● Other yeasts

The genomic approach to analyse *Sac. cerevisiae* provided the impetus for a similar genome sequencing project for *Sch. pombe* which is expected to be completed later this year. The second microbial eukaryote to be sequenced will provide important comparative information on these two brewing yeasts with a unicellular life form.

There are over 800 yeast species described in the latest taxonomic treatise but few have been studied genetically. Those that have possess interesting metabolic activities

making them of potential economic importance. For example, *Kluyveromyces lactis* can utilize cheese whey, a waste product of the cheese industry, several *Pichia* species can grow on methanol, *Yarrowia lipolytica* and *Candida maltosa* can utilize hydrocarbons as carbon sources, *Debaryomyces (Schwanniomycetes) occidentalis* can grow on starch, *Arxula adenivorans* can utilize nitrate, and *Pichia guilliermondii* can over-synthesize riboflavin. Developing genetic systems – the ability to do crosses and have appropriate vectors (such as plasmids) to propagate genes independently of the chromosomes – is very time-consuming. However, genomics can provide an alternative route around these problems. Direct sequencing and making homology comparisons can identify genes of interest. Furthermore, it is often easy to express and analyse a gene from one organism in another, so-called ‘surrogate’ genetics. A good example is the naturally diploid yeast *Candida albicans* (Fig. 3). Neil Gow, Al Brown and Duncan Shaw have made Aberdeen a major international centre for the genetic and genomic analysis of this genetically intractable yeast. They form part of an international consortium that is sequencing the entire genome, due to be available next year. The results will undoubtedly provide valuable information to devise new strategies to attack this important human pathogen, which is a major killer of immunocompromised patients.

● The future

Although founded in the 19th century, genetics is essentially a 20th century science and genomics is its 21st century successor. *Sac. cerevisiae* and *Sch. pombe* will continue to remain model organisms for some time to come, but the full potential of yeasts has not yet been realized. The gene resources found in yeast species could be used for improvements in making bread, wine, beer and fuel ethanol, and in many other ways. However, public perception of the conjectured hazards of GMOs suggests that all microbiologists will need to work hard to convince consumers of the potential benefits of organisms we are now able to understand and exploit as never before.

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ABOVE:

Fig. 2. Part of a *Saccharomyces cerevisiae* micro-array. The colour and intensity of the spots give information on the amount of RNA hybridized to the DNA spot.
COURTESY PROFESSOR PATRICK BROWN, STANFORD UNIVERSITY, USA

LOWER LEFT:

Fig. 3. *Candida albicans*. Filamentous forms of this human pathogen containing an engineered green fluorescent protein gene glow brightly on infected murine kidney cells (red).
COURTESY PROFESSOR NEIL GOW, UNIVERSITY OF ABERDEEN

Further reading

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Yeast Virtual Library website: <http://genome-www.stanford.edu/Saccharomyces/VL-yeast.html>

