The 'lab on a chip' and its impact on drug discovery

Few technology developments have had as profound an effect on society as the invention of the microchip integrated circuit in the 1960s. Since then, the power of electronic circuits has increased while their cost in real terms has dropped, and both by similar proportions. The evolution of microprocessors gives an excellent illustration of this with Moore's law[1] providing a prediction which has proved remarkably accurate to date. In fact, information technology is the key area to which microchip technology has been applied, and its influence has been immense, not least on science. Miniaturisation has gone hand-in-hand with this, and it is now one of the most important technology developments of our times. Most visible in electronics, computing and telecommunications, miniaturisation has revolutionised everyday life, and personal computers and telephones, whose power is vast compared to their 1970s counterparts, are now household items.

by Dr Simon Cowen and Dr Coulton Legge

In recent years, those involved in chemistry, biochemistry and analytical science have sought to use microchip technology to stimulate a revolution akin to that undergone by electronics, by applying it to miniaturise the procedures and techniques presently carried out in the laboratory. The driving factors behind this are many and they include, as well as the opportunities for carrying out novel science, significant and growing commercial pressures to reduce costs and the need for ever growing sample throughput. The pharmaceutical industry has been one of the key drivers behind this, and for a number of reasons which we will outline in this article. The result of this scientific and commercial interest is the 'lab on a chip', a term which has been coined to describe either the reduction in size of current laboratory techniques to a form which offers faster, more cost-effective processes (measurement and synthesis for example); or the development of new methods of processing smaller quantities of sample which are faster and more efficient than their laboratory-based counterparts. Chip technology is the factor common to both approaches, and in this article we describe how it offers a way of meeting current challenges in drug discovery, and how it is predicted to affect drug discovery over the coming decade.

Technology overview

Electronic circuits are made using microfabrication techniques[2], and it is these which have been adapted to the design and production of lab on a chip devices. The basic principles are not unlike printed circuit board methods, in that the circuit pattern is transferred to a suitable substrate (glass or silicon for example) by means of photosensitive chemicals called resists. By then etching or building up material it is possible to

The future - microfluidics offers the possibility of building an entire laboratory in the palm of the hand

Photograph courtesy of LGC

define and generate physical structures on a microscopic level. Further bonding (of a lid to enclose channels for example), coating and etching steps turn this structure into a working device with features from millimetres to below 1 micrometre in size. Chemical and biochemical applications of microfabrication include separation channels, detectors, pumps, valves, reactors, DNA hybridisation chips, and a multitude of combinations thereof[3]. All of these have been fabricated separately, and the challenge now is to integrate the individual components into complete, self-contained systems to form the ubiquitous 'lab on a chip'.

The first published attempts to use chip technology in the miniaturisation of commonly used analytical techniques were for gas and liquid chromatography, in 1975 and 1990 respectively. Although they showed promise, technical difficulties in producing a complete, integrated design prevented commercial exploitation and further development. The breakthrough came about 10 years ago, when a group at what were then Ciba-Geigy's research laboratories, while investigating novel approaches to chemical analysis, demonstrated how capillary electrophoresis could be carried out on a glass chip[4]. Similar work on this and related techniques gave rise to the new field of microfluidics, which is now a strong growth area. At the same time, another application of microchips for rapid measurement was conceived, with the proposal of DNA chips for genetic analysis being proposed and demonstrated[5]. The two approaches have since grown into a new science which has expanded and matured to the extent that it now supports many conferences and commercial interest is strong.

Applications in drug discovery

Bringing a new drug to market is an expensive and time-consuming business – R&D costs have grown rapidly over the last decade or so, and current estimates put the cost of developing a new product at roughly $500 million, over some 10-15 years[6]. This rise in costs and the competitive nature of the pharmaceutical industry are continually forcing the process of identifying lead compounds to be more efficient, and the result is that greater numbers of compounds must be screened more quickly and in smaller quantities. At the same time, as the introduction of new medicines is more tightly regulated, and the time to patent expiry is finite, these R&D costs must be recovered while keeping drug prices reasonable.

The result has been a technology shift towards combinatorial and high-throughout chemistry, which increase chemical diversity by producing larger sets of compounds, and allow the lead discovery process to cover a larger chemical range. The key technologies that have enabled this are automation and miniaturisation, the latter beginning with the introduction of the 96-well assay plate (since extended to 384 and 1536 wells). With the latest automated robot-operated assay platforms, it is now possible to screen thousands of compounds in one day.

However, the introduction of pharmacogenomics to drug design, using the increasing knowledge base of how genetic factors affect drug action on the body and individuals' response to those drugs, will mean that new products will increasingly be developed from the starting point of a particular gene or group of genes. Since the data sets generated will be so much larger, the trend towards miniaturisation will become even more important, but it is recognised that there is a limit to what can be achieved with current technology. For example, with size reduction come new problems such as how to handle small volumes of liquid, evaporation in open wells, and so on.

Chip-based solutions provide the means to circumvent these problems, and to extend measurement and production capability by working in a new (the microfluidic) regime. Firstly, the use of systems designed specifically for handling nanolitre volumes in a small space allows parallel processing on a large scale. Secondly, microfluidics can exploit the different properties of fluids at the microscale to make reactions and measurements more efficient (mixing, fluid flow and reaction chemistry are all potentially more effective at smaller dimensions). With this in mind, given the pressures being placed on the drug discovery process, 'lab on a chip' will be crucial to success in a number of ways:

Genetics research

With the first draft of the human genome, and the 30,000 or so distinct genes which have already been identified, the task is now to relate these genes, and individual genetic variations such as the single nucleotide polymorphisms (SNPs) which occur along the genome, to individuals' response to preventative and therapeutic medicines. This is a huge task which involves not only the screening of known gene sequences for activity, but also the identification of new genetic information. Development of the tools needed to do this is attracting large-scale investment and research effort, and has resulted in some novel technologies. One of the most significant of these is the DNA microarray[5], which works by providing a matrix of thousands of oligonucleotides to which a target is exposed, the subsequent hybridisation being detected, indicating which genes are active. Microarray assays provide a much faster method than the traditional membrane assays, and are generally recognised to have contributed to the early appearance of the first draft of the human genome. Other technologies based on parallel microfluidics are also under development, for DNA, RNA and protein measurement. The enhanced speed, reduced sample volumes and amenability to parallel, high-volume analysis of these technologies offer ways of breaking current limits of throughput.

Genomics gives information about which proteins will be expressed under a given set of conditions, and the code for a particular protein is determined by a DNA sequence. Since disease and treatment work at the protein level, profiling of healthy and diseased tissues will reveal much more information than genomics can on its own. Current effort is therefore increasingly turning towards proteomics, which aims to study all of the proteins in a cell and how they interact with each other. This is an even more demanding task than genomics, since there are so many more possible protein interactions to be considered, and the need for instrumentation which can deliver the amounts of data involved is even more acute.

High throughput measurement and synthesis

As the requirement grows for synthesising and analysing larger numbers of compounds for lead

identification and optimisation, conventional instrumentation will reach the limit of its capability, as it will with genetics research. Lab on a chip methods based on parallel microfluidics are being developed to overcome not only this, but also to allow larger quantities for subsequent clinical testing by scaling out production (parallel synthesis) rather than scaling up. Throughput is also improved with the increased speed at which miniaturised analysis can deliver results, and costs lowered with reduced reagent quantities and power consumption.

Response to candidate drugs

Once lead compounds have been identified and pass on to become candidate drugs, the body's response has to be determined, to eliminate side effects such as genetic or metabolic reactions, and to screen out the unsuitable candidates. In order to make this process more efficient, improved *in vitro* and *in vivo* screening methods are needed – methods that are both faster and of sufficient robustness to satisfy the regulatory conditions which are in force.

Informatics

Given the increased throughput that miniaturisation offers, it is not surprising that the amount of data generated has increased greatly, and it all has to be interpreted and sorted. The result is the emergence of bioinformatics, which seeks to extract useful information from a large background of data and to develop sophisticated algorithms to uncover relationships between genes, SNPs and disease. Miniaturisation is again essential as information technology becomes more powerful yet cheaper in real terms, and software of the required complexity can be developed.

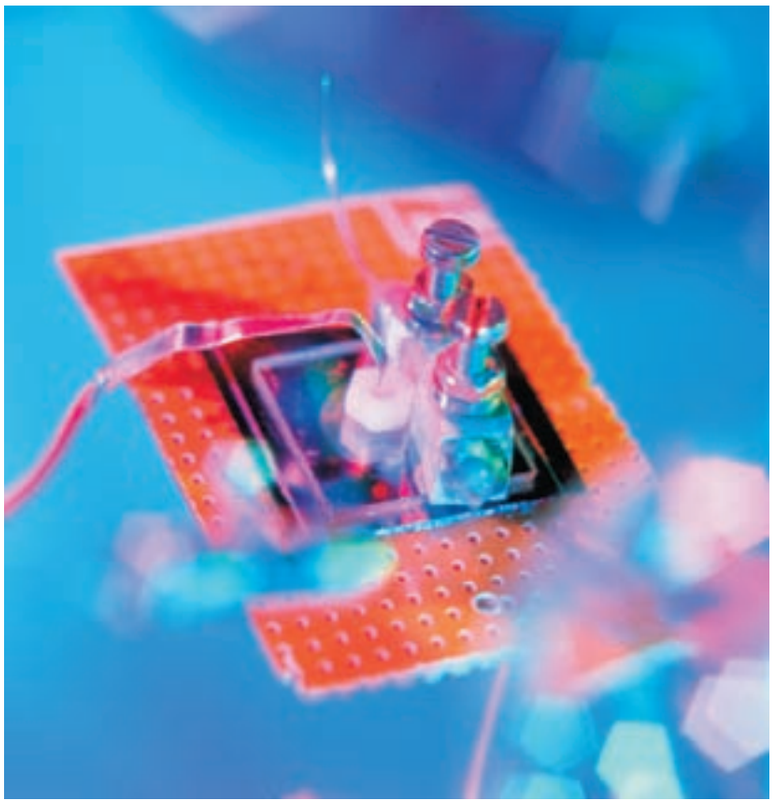
Technology suppliers

Commercial development of chip-based chemistry has principally been centred around hi-tech start-up companies formed to exploit the work of academic research groups, and the involvement of large end users in strategic alliances and/or large-scale shareholdings. Several technology options are available, primarily for research at this stage, which can be broadly grouped into two categories:

Microfluidics

Conveniently defined as the handling of liquids in spaces of micrometre dimensions, microfluidics offers both a means of miniaturising standard laboratory methods and of controlling liquid samples in new ways. Pressure and electrokinetic methods are the principal means of driving fluids around microchannel networks, although at least one manufacturer offers a platform based on a spinning CD arrangement[7]. The problem of generating high pressures on a chip also appears to have been solved recently, with the development of an electrokinetic high-pressure pump[8]. Of particular interest in microfluidics is the different behaviour patterns that liquids exhibit in this size regime (for example mixing behaviour, flow profiles and reaction chemistry), the central theme being the identification of which of these can be used to advantage in novel processes.

To date, microfluidics has been incorporated into several instruments, notably chromatography and electrophoresis, to improve performance and reduce footprint (either for use in the laboratory or in a mobile role). Agilent's Bioanalyzer[9] is the first complete product in which microfabrication is used to construct the heart of the system, but microfluidic chips themselves are available from a variety of vendors. Similar instruments for DNA amplification and analysis, and other separation



Photograph courtesy of LGC

An on-chip chromatography system

References

1 In 1965, the co-founder of Intel Corporation, Gordon Moore, predicted that a new version of a given microprocessor would be released every 12-18 months, have double the computing power of its predecessor, at approximately the same cost.
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5 Schena, M, Shalon, D, Davis, RW and Brown, PO. Science, 270 (1995) 467-470.
6 www.phrma.org
7 www.gyrosmicro.com
8 www.eksigent.com
9 www.chem.agilent.com

Links

1 www.labonachip.org.uk, The UK Lab on a Chip Consortium.
2 www.lab-on-a-chip.com, Commercial news and links.
3 www.calipertech.com, Caliper Technologies, Microfluidics.
4 www.aclara.com, ACLARA Biosciences, Microfluidics.
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6 www.affymetrix.com, Affymetrix Corporation, DNA chips.
7 www.nanogen.com, Nanogen, DNA chips.
8 www.ogt.co.uk, Oxford Gene Technology, DNA chips.
9 www.micralyne.com, Micralyne, Design/fabrication.
10 www.gene-chips.com, Resources on genome chips.
11 www.chipstohits.cm, Annual Chips to Hits conference.
12 www.rsc.org/loc, new 'Lab on a Chip' journal.

methods are also under development, but the true microfabricated, integrated 'lab on a chip' is still some way off.

Microarrays

Array platforms based on several construction methods are available for research purposes, including photolithography, mechanical spotting, electronic addressing as well as others equally novel. Two chip sizes, the typical microscope slide and the Affymetrix cartridge, are emerging as standards in the industry, and this is expected to aid development in the same way that hardware standardisation did for the PC industry. As DNA arrays become essential in drug discovery, the move towards proteomics is stimulating the development of the protein array, which is still very much at the initial conception stage. Dealing with proteins on a chip is much more complex, with factors such as the larger size of the human 'proteome', stability and protein structure to be considered.

Conclusions and outlook

Microchip technology in chemistry and biochemistry has now reached the stage where commercial products are a reality and are beginning to be adopted in the pharmaceutical industry. Although the promise originally predicted 10 years ago has some way to go before being fulfilled, and there are some complicated patent disputes to be settled, the adoption of microchip technology by industry has begun and is set to continue.

There are still some technology issues to be resolved, one of the more significant of these being initial sample handling and preparation, as most platforms work with samples prepared using standard methods. In some situations this may represent a bottleneck which will prevent throughput rising to the levels required for discovery based on the new approach of genetics and proteomics. Integration with the laboratory, whether to another instrument such as a mass spectrometer, or the outside world, is also key as it defines to some extent the design approach, be it single chip or separate connected elements in one package. Standard accepted formats will also be needed, to ensure that components will fit together and there is strong interchangeability between manufacturers. The large amounts of information generated dictate that steps should be taken to ensure that errors are not introduced into either the production or data capture stages, so that the information obtained is reliable. Despite these considerations, the rate of uptake of microchip technology is strong in the pharmaceutical industry, and it is likely that the biochip market will eventually realise its potential. DDW

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