Institution:
University of Glasgow

Unit of Assessment:
B15: General Engineering

Title of case study:
Gene Sequencing on Silicon: the Ion Torrent Personal Genome Machine

1. Summary of the impact

The development of microelectronic sensor arrays for biological applications, pioneered at the University of Glasgow, is central to a unique gene sequencing system developed by Ion Torrent. The Ion Torrent personal genome machine is a bench-top system that, compared to optically mediated technologies, is cheaper and easier to use. Ion Torrent was founded in 2007 and bought by Life Technologies in 2010 for $725M; they, in turn, were bought by Thermo Fisher for $13Bn, citing Ion Torrent as a motivation. Ion Torrent now has 62% of the bench-top sequencing market, estimated to be worth $1.3Bn in 2012.

2. Underpinning research

The Microsystem Technology Group at the University of Glasgow was set up by David Cumming in 1999 with the aim of combining microelectronics technology with sensors and nanofabrication to create physical, biological and medical sensors. A major objective has been to exploit commercially available **complementary metal oxide semiconductor** (CMOS) technology by new design, augmentation, and demonstration in non-traditional applications. In 2001 the group won a Scottish Higher Education Funding Council Research Development Grant (RDG) to work on Integrated Diagnostics for Environmental and Analytical Systems (IDEAS, RDG 131). The project was a collaboration between the Universities of Glasgow and Edinburgh, led by Cumming (Lecturer 1999-2001, Senior Lecturer 2001-04, Professor 2004-present). A key pilot device for IDEAS was the laboratory-in-a-pill, for which considerable effort into miniaturisation was required.

With this motivation, work began in 2001 on the implementation of **ion sensitive field effect transistors** (ISFETs) in foundry CMOS integrated circuits. ISFETs are used for measuring ion concentrations in solution. Cumming collaborated with Philips through a CASE award (Paul Hammond, Research Student 2000-04, PDRA 2004-06, Lecturer 2006-07) and was able to demonstrate fully CMOS compatible ISFETs [1]. ISFETs can be made selective for several different ion species, and it is beneficial that silicon nitride, a standard passivation material used in CMOS, renders ISFETs selective to hydrogen ions. Because of this, the team was then able to demonstrate the world’s first fully instrumented digital pH meter on a single chip, fulfilling substantially one of the miniaturisation requirements of IDEAS [2]. This level of sophistication proved that ISFET **sensor system on chip** was feasible.

In 2001 Cumming was awarded an EPSRC Advanced Research Fellowship (Scalable Nanotechnology for Integrated Sensor Arrays, GR/A10987/01) to work on integrated circuit technologies and their application to biological sensing. Following on from the Philips CASE award, Cumming’s focus with support from Hammond and Mark Milgrew (Research Student 2001-05, PDRA 2005-08) was integration of ISFETs into a scalable and addressable CMOS architecture. This was initially demonstrated with a very small 2 x 2 array of ISFETs [3]. As a next step, Cumming’s group worked on increasing the array size, overcoming technical challenges in the electronic implementation to improve the signal transduction from the array [4]. The result was a 16 x 16 sensor array, adequate for experiments with cell tissue culture on the CMOS chip itself. The design was scalable, meaning that simple replication could be used to expand the array size to millions of sensors, and that each individual sensor could be miniaturised by progressing to smaller geometry CMOS technologies. This innovation was central to the success of Ion Torrent.

In 2006 Cumming was awarded an EPSRC Science and Innovation Award as PI to set up an Electronics Design Centre for Heterogeneous Systems (EP/D501288/1). Further work on refining the CMOS ISFET sensor array led to its first practical demonstration as a biological sensor in collaboration with Dr Mathis Riehle (Research Assistant 1996 – 2000, Lecturer 2000-2005, Reader 2006-present, Centre for Cell Engineering). The work was inspired by CMOS focal plane array designs, thus the chip was known as the proton camera.

This research was read by Jonathan Rothberg (the founder of 454 Technologies), who filed his first patent for Ion Torrent in 2007, citing Cumming's papers as prior art. With the sensor arrays central to the Ion Torrent system, Rothberg approached Cumming directly for his expertise. The work was then presented at the prestigious International Solid State Circuits Conference in 2008 – a leading microelectronics forum [5]. A significant new result at this time was the demonstration of optical annealing to homogenise the otherwise disparate threshold voltage characteristics of the so-called "floating-gate" sensors in the array. Without this insight the development of large arrays with sufficiently close tolerance between the devices would not be achieved [6]. Well-matched devices are necessary for Ion Torrent since ideally all the sensors should be as close to identical as possible. This later work was also cited in follow-on patents and journal articles by Ion Torrent.

3. References to the research

[1] Hammond, P.A., Ali, D., Cumming, D.R.S., Design of a single-chip pH sensor using a conventional 0.6 μm CMOS process, *IEEE Sensors Journal*, vol. 4, no. 6, 706-712, Dec 2004. DOI: 10.1109/JSEN.2004.836849

[2] Hammond, P.A., Ali, D., Cumming, D.R.S., A system-on-chip digital pH meter for use in a wireless diagnostic capsule, *IEEE Transactions Biomedical Engineering*, vol. 52, no. 4, 687-694, Apr 2005. DOI: 10.1109/TBME.2005.844041 *

[3] Milgrew, M.J., Hammond, P.A., Cumming, D.R.S., The development of scalable sensor arrays using standard CMOS technology, *Sensors and Actuators B*, vol. 103, no. 1-2, 37-42, Sept 2004. DOI: 10.1016/j.snb.2004.03.004

[4] Milgrew, M.J., Riehle, M.O., Cumming, D.R.S., A large transistor-based sensor array chip for direct extracellular imaging, *Sensors and Actuators B*, vol. 111(SI), 347-353, Nov 2005. DOI: 10.1016/j.snb.2005.01.020

[5] Milgrew, M.J., Riehle, M.O., Cumming, D.R.S., A 16x16 CMOS Proton Camera Array for Direct Extracellular Imaging of Hydrogen-Ion Activity, *IEEE Solid-State Circuits Conference, Feb 2008, Digest of Technical Papers* pp. 590-638, 2008. DOI: 10.1109/ISSCC.2008.4523321 *

[6] Milgrew, M.J., Cumming, D.R.S., Matching the transconductance characteristics of CMOS ISFET arrays by removing trapped charge, *IEEE Transactions Electronic Devices,* vol. 55, no. 4, pp. 1074-1079, Apr 2008. DOI: 10.1109/TED.2008.916680 [REF2] *

* best indicators of research quality

4. Details of the impact

Genetic sequencing stirred worldwide attention in the 1990s with the human genome project. However, work to identify and map the whole of the human genome proved to be slow and laborious. Two companies that started up to capitalise on the opportunity were Illumina (1998) and 454 Technologies (1999), who created technologies based on optical systems. Jonathan Rothberg, the founder of 454 Technologies, went on to establish Ion Torrent Incorporated in 2007 with the aim of developing a system based on arrays of sensors in an integrated circuit to eliminate the complexity of an optical system. His objective was to produce a small bench-top "next generation sequencing" system that was cheap and comparatively easy to use. Today, bench-top sequencing is entirely dominated by two products: Ion Torrent Personal Genome Machine; and MiSeq from Illumina. Recent data showed the market in 2012 to be worth $1.3Bn in sales, with projected growth to $2.7Bn by 2017. In 2012 Ion Torrent had captured 62% of the market and dominates sales into diagnostic laboratories owing to its low cost, speed and ease of use.

Impact of Ion Torrent technology on research, development and investment
Professor Cumming at the University of Glasgow was first contacted by Jonathan Rothberg in 2007 specifically because he had developed the technology for implementing large arrays of sensors on CMOS for hydrogen sensing and demonstrated that massive scaling was possible. Furthermore, a major obstacle to the development of new sequencing techniques had been that all the sensors must lie within a tolerance band: Cumming's research had made it possible for ISFETs to meet this tolerance criterion. As a consequence of the research done at the University of Glasgow, DNA hybridisation reactions on a microelectronic chip could be used for sequencing to achieve the objectives of Ion Torrent.

Ion Torrent technology is variously referred to as post-light sequencing, semiconductor sequencing, and "the chip is the machine", reflecting the pioneering use of microelectronic chips for DNA sequencing. Many of the University of Glasgow research papers are serially cited [1-6] in at least 12 patents that are essential to the Ion Torrent technology, wherein the research is described as "exemplary". In 2008 Mark Milgrew, who had, as a research student and PDRA, worked with Cumming, was hired by Ion Torrent to work on their chip design. Furthermore, Cumming was also hired under contract as a member of the Scientific Advisory Board to advise on CMOS array technology. Ion Torrent proceeded to develop the technology in "stealth-mode" until beta releases to selected test sites were made in late 2009. The introduction of this disruptive technology led to the rapid sale of Ion Torrent to Life Technologies in August 2010 for $725M. Detailed disclosure of the technology was not published until 2011 [Rothberg *et al*, Nature, July 2011].

Life Technologies continued to develop the technology, leading to successive releases of their sequencing chips for use in the Ion Torrent machine. The systematic increase in sensor numbers has led to chips with close to 1Bn ISFETs (the Ion Proton system). The Proton is aimed at the whole genome market whereas the Ion Torrent is targeted toward short sequences. Thermo Fisher acquired Life Technologies in 2013 for $13Bn, citing the acquisition of the Ion Torrent/Proton as one of the motivations.

Impact of Ion Torrent technology on detection of diseases, mutations
Ion Torrent technology, enabled by the pioneering research of Cumming and his team at the University of Glasgow, is especially suited to reading long sequences from small genomes or targeted sections of larger genomes. It is therefore excellent for diagnostic and medical research applications and is widely used to allow confident detection of DNA mutations responsible for specific diseases including breast, bowel, and lung cancer. Searches can also be made for genetically passed on disorders including cystic fibrosis and Duchenne muscular dystrophy.
The Ion Torrent Personal Genome Machine also allows research labs to look at RNA (the intermediate stage between genes and proteins) to determine how they change in different circumstances e.g. in disease. This develops understanding of disease mechanisms, helping the development of new therapies for e.g. myotonic dystrophy, oral and skin cancers, and Alzheimer's. For all these applications, where only small amounts of genetic data are required, Ion Torrent is about one third of the cost to use compared to its competitors and this is a contributing factor to its market dominance.

Ion Torrent devices particularly lend themselves to sequencing of small genomes, e.g. bacterial genomes. An excellent and highly publicised example is the *E. coli*. O104:H4 outbreak in Germany in 2011 that was responsible for 50 deaths. Ion Torrent sequenced the bacterium in a day, leading to targeted drug therapy for those affected. A further 4000 people were diagnosed and treated successfully. This was discussed in a BBC news broadcast (14 June 2011) to which Cumming contributed.

5. Sources to corroborate the impact

- Example patent. This is the first Ion Torrent patent and it cites the reliance of Ion Torrent's IP on research at the University of Glasgow
    - US Patent 7948015, Rothberg, J.M. et al, 14 Dec. 2007. (Available from HEI)
- Further examples of patents citing University of Glasgow research

   - US Patent 8263336, Rothberg, J.M. et al, 31 May 2011. (Available from HEI)
   - US Patent 8247849, Fife, K., Johnson, K. Milgrew M., and 15 Mar. 2012. (Available from HEI)
   - US Patent 8217433, Fife, K., and 15 Mar. 2012. (Available from HEI)
- Website documenting sale of Ion Torrent to Life Technologies
   - https://ir.lifetechnologies.com/releasedetail.cfm?ReleaseID=520347
- Website documenting sale of Life Technologies to Thermo Fisher
   - http://news.thermofisher.com/press-release/corporate/thermo-fisher-scientific-acquire-life-technologies-corporation
   - http://uk.reuters.com/article/2013/04/15/us-lifetechnologies-thermofisher-idUSBRE93D0A620130415
- Ion Torrent product website describing their main applications
   - http://www.lifetechnologies.com/uk/en/home/life-science/sequencing/next-generation-sequencing/ion-torrent-next-generation-sequencing-applications.html
- Website providing recent sales data
   - http://www.bio-itworld.com/news/02/18/13/Ion-Torrent-edges-Illumina-sales-benchtop-sequencers-Macquarie.html
- BBC News story on Ion Torrent featuring David Cumming
   - http://www.bbc.co.uk/news/uk-scotland-glasgow-west-13756119
- Website comparing the cost of sequencing for various technologies. Please note distinction between large volume sequencing, and the sequencing of short strands. Depending on which chip is used, Ion Torrent is competitive in both markets.
   - http://nextgenseek.com/2012/08/comparing-price-and-tech-specs-of-illumina-miseq-ion-torrent-pgm-454-gs-junior-and-pacbio-rs/

(Copies of above web-based content also available from HEI)

Testimonial (available from HEI)
Founder of Ion Torrent Systems Incorporated confirming that University of Glasgow research proved it was possible to make arrays of ISFECT in a commercial foundry, enabling mass manufacture at low cost and thus making it possible for Ion Torrent to develop their sequencing devices.