Institution: University of Cambridge		
Unit of Assessment: 8		
Title of case study: Sphere Fluidics		
Period when the underpinning research was undertaken: 2005-present		
Details of staff conducting the underpinning research from the submitting unit:		
Name(s): Professor Chris Abell	**Role(s) (e.g. job title):** Professor of Biological Chemistry	**Period(s) employed by submitting HEI:** 1984-2020
Period when the claimed impact occurred: 01/08/2013-31/07/2020		
Is this case study continued from a case study submitted in 2014? N		

1. Summary of the impact

Sphere Fluidics is a spin out company that was set up to exploit microdroplets technology developed in the Department of Chemistry at the University of Cambridge. The use of microdroplets as small scale, high throughput reactors is at the heart of their new instrument for the automated generation and selection of monoclonal antibodies, the Cyto-Mine®. Launched in 2018, this instrument is already being used in the biopharmaceutical industry for the development of new antibody therapies. At the end of the 2019 financial year Sphere Fluidics had 31 employees [text removed for publication], and this year they expanded to open a new facility in Cambridge as well as a US office. In addition to instrument development, the company is the major worldwide supplier of the hardware and consumables required to work with microdroplets.

2. Underpinning research

In 2005 Professor Chris Abell and Prof Wilhelm Huck in the Department of Chemistry at the University of Cambridge set up a group together with Dr Florian Holfelder in the Department of Biochemistry to develop microdroplet technology. The Cambridge Microdroplets group have made a number of important fundamental contributions to microdroplets methodology and published over 70 papers in the area since 2005. Microdroplets show great promise as a new high-throughput technology in chemistry, biochemistry and molecular biology.

Microdroplets. The technology involves generating water droplets 30-100 µm in diameter that are pumped in a fluorocarbon carrier oil inside a microfluidic system. A key part of the technology is the use of bespoke surfactants that make the droplets exceptionally stable, allowing them to be used as microreactors for chemistry or biology on a very small scale. The droplets are generated at a rate of 1,000-10,000 per second, and any reactions that take place inside are monitored by changes in the fluorescence of the contents. The use of microfluidics to manipulate the droplets enables sorting of droplet populations and merging of specific droplets in a highly controlled manner.

The Cambridge Microdroplets group contributions in the area of method development include new hardware and new reagents: the development of polydimethylsiloxane mixers to fuse droplets; the use of special traps for monitoring changes in the content of an individual droplet over time; synthesis of novel tri-block surfactants based on fluorous oils that form exceptionally stable droplets. On the applications side, they have also made a number of important contributions: they developed a device for in-droplet PCR amplification of DNA; they developed reagents and a protocol for *in vitro* transcription and translation; they were able to simultaneously monitor protein expression in bacterial cells and the activity of a secreted enzyme.[R1] They were the first to show that microdroplets could be used to monitor changes in microorganism growth and metabolism over time, and used this methodology to study algal development and the production of lipids and ethanol by these organisms.[R2] A major technical focus was the development of an alternative to fluorescence spectroscopy to characterise the contents of microdroplets. The Cambridge Microdroplets group pioneered the use electrospray ionisation mass spectroscopy (ESI-MS) to monitor droplet contents. Most significantly they showed that ESI-MS could be used to study

proteins in droplets, removing the need for fluorescence labelling, and opening up microfluidic technology to the study of native proteins.[R3,R4]

Exploitation. Realising the potential of their discoveries, the group quickly patented the key technology for accessing, analysing and handling the microdroplet contents.[R5–R7] In 2010, Abell and Huck co-founded Sphere Fluidics to exploit microdroplet technology. The company was initially based in the Department of Chemistry and moved to the current premises on the Babraham Research Campus in 2013. Abell has chaired the Scientific Advisory Board from the outset.

3. References to the research

R1. Shim, J.-U.; Olguin, L. F.; Whyte, G.; Scott, D.; Babtie, A.; Abell, C.; Huck, W. T. S.; Hollfelder, F. Simultaneous Determination of Gene Expression and Enzymatic Activity in Individual Bacterial Cells in Microdroplet Compartments. *J. Am. Chem. Soc*. **2009**, *131*, 15251-15256.

R2. Huebner, A.; Srisa-Art, M.; Holt, D.; Abell, C.; Hollfelder, F.; DeMello A. J.; Edel, J. B. Quantitative Detection of Protein Expression in Single Cells Using Droplet Microfluidics. *Chem. Commun.* **2007**, 1218-1220.

R3. Fidalgo, L. M.; Whyte, G.; Ruotolo, B. T.; Benesch, J. L. P.; Stengel, F.; Abell, C.; Robinson, C. V.; Huck, W. T. S. Coupling Microdroplet Microreactors with Mass Spectrometry: Reading the Contents of Single Droplets on Line. *Angew. Chem. Int. Ed.* **2009**, *48*, 3665-3668.

R4. Smith, C.; Li, X.; Mize, T. H.; Sharpe, T. D.; Graziani, E. I.; Abell, C.; Huck, W. T. S. Sensitive, High Throughput Detection of Proteins in Individual, Surfactant Stabilized Picoliter Droplets using NanoESI Mass Spectrometry. *Anal. Chem.* **2013**, *85*, 3812-3816.

R5. Abell, C.; Huck, W. T. S.; Bratton, D.; Whyte, G.; Fidalgo, L. M. Microfluidic Systems. US2012091004 (A1), 2012.

R6. Abell, C.; Huck, W. T. S.; Sharpe, T.; Smith, C.; Mize. T.; Robinson, C.; Li, X. Ionisation Mass Spectrometry. US20130187040 (A1), 2013.

R7. Abell, C.; Huck, W. T. S.; Craig, F. F. Method of Providing a Chemical or Biological Material in a Quantised Form and System Therefor. US2013139477 (A1), 2013.

Research outputs published in peer-reviewed journals.

4. Details of the impact

Antibody therapeutics. Seven of the top ten selling drugs on the market are monoclonal antibodies, and according to Fortune Business Insights in 2019 the monoclonal antibodies therapy market was worth USD123,030,000,000. However, the discovery of new antibodies with beneficial properties is a process which is complex, expensive and time-consuming. It was clear that antibody discovery was an area where microdroplets could have a significant impact through automation of the parallel screening required. Sphere Fluidics have used the microdroplet technology developed in the Department of Chemistry by the Cambridge Microfluidics group to develop an instrument for the automated discovery of therapeutic monoclonal antibodies, which is set to transform the way that the biopharmaceutical industry works in this area.

Sphere Fluidics. The company was initially spun out as a platform technology company to develop commercial opportunities using microdroplets to address industrial challenges through partnerships with pharmaceutical and biotech companies.[E1] Capital investments in the company have reached a total of GBP17,000,000 and has 325 international customers. At the end of the 2019 financial year the company had 31 employees [text removed for publication].[E2] The company recently expanded to a second site at Granta Park in Cambridge and also has a US office at Monmouth Junction (NJ). The major success of the company has been the development of a range of new single cell analysis systems for biotherapeutic discovery (see below), and these achievements were recognised with a Queen's Award for Enterprise for Innovation in 2020.[E3]

The Cyto-Mine®. Sphere Fluidics initially assisted collaborators in the pharmaceutical industry to develop microdroplet capability in house by building bespoke research platforms from commercially available lasers, pumps, and microscopes. However, it was clear that to move the adoption of the technology to the next stage, a desktop instrument was needed that could be

operated by technicians. Sphere Fluidics therefore developed the Cyto-Mine®, which was launched in 2017.[E4] This instrument allows up to 40,000,000 discrete antibody-producing cells to be individually encapsulated in microdroplets. On incubation, each cell secretes a single antibody into an individual microdroplet. A fluorescence-based binding assay is then used to quantify the activity of the antibodies contained in each droplet. The user uses the automated binding assay to select the most promising microdroplets, and the instrument dispenses each distinct member of this subset of droplets into individual wells of a microtiter plate. Monoclonality is confirmed from video capture of each droplet. By collecting individual droplets, individual cell lines are selected, which can then be grown up to obtain the active monoclonal antibody on scale. This powerful new technology outperforms all existing technologies by sorting, imaging, and dispensing in a single platform, and it has already won a number of awards, including the Frost & Sullivan European New Product Innovation Award.[E5] Traditional methods in this field require different types of equipment for each step, which increases costs, turnaround time, and the risk of sample contamination. These can also lead to ineffective data processing and data reproducibility issues. The Frost & Sullivan analysis found that the Cyto-Mine® is significantly faster than competitor technologies and that monoclonality is considerably better (99.9% compared with 95-99%), all for USD450,000 per instrument. They conclude that "*Cyto-Mine® is disrupting the therapeutics and diagnostics industry by outperforming traditional technologies; so new biopharmaceuticals can be produced faster with improved monoclonal antibody screening and more efficient research*".[E5] The Cyto-Mine® was also ranked number one in The Scientist Top 10 Innovations. Dr T Kelly tested the Cyto-Mine® at Janssen and found that a major advantage is that the instrument provides proof of clonality, i.e. confirmation that there is a single, verified cell in each well. This feature has allowed simplified protocols which half the processing time: "*the time it takes to go from transfection to freezing vials in the old process would have been three months*, *and now it's six weeks*", Kelly claims.[E6]

Sphere Fluidics have entered into partnerships with Oxford Genetics (in 2019) and Geneva Biotech (in 2017), which has provided funding (EUR1,600,000) to adapt the Cyto-Mine® to make automated, benchtop devices for gene editing.[E7,E8] Current gene editing technologies are effective at creating small local gene modification, but are severely limited by a lack of tools to dock complex multigene circuits into defined genomic sites. This new adaptation of the Cyto-Mine® has applications in gene therapy, cancer immunotherapy, stem cell reprogramming, and drug discovery.

The ESI-Mine™. Sphere Fluidics have developed another desktop instrument, ESI-Mine™, the first high-throughput, miniaturised, electrospray injection mass spectrometry product that can be used to screen reactions in microdroplets.[E9] Compared to current technology, the ESI-Mine™ is not only faster (0.4 seconds per sample compared to 5-10 seconds), but can also handle more than 10 times the number of samples per day (more than 200,000 compared with 8,000-16,000).[E9] ESI-Mine™ was recognised as the most innovative single cell analysis platform in the 2018 Global Health and Pharma Biotechnology Awards.[E10]

Wider product portfolio. In addition to new instruments, the company designs and sells all of the key components required for microdroplet research: microfluidic instrumentation (Pico-Gen™ and Pico-Safe™) and a range of proprietary chemical products (Pico-Surf™, Pico-Break™, Pico-Glide™ and Pico-Wave™).[E11] Sphere Fluidics have now signed up 11 distributors to sell their products globally, so that most microdroplets groups in the world whether in industry or academia use Sphere Fluidics products.[E12]

5. Sources to corroborate the impact

[E1] Sphere Fluidics – Company website 31.07.2019. "About Us"

[E2] Letter from CEO and Company Director of Sphere Fluidics 03.12.2020

[E3] BusinessWire – Queen's Award for Enterprise 29.04.2020. "Sphere Fluidics Receives Queen's Award for Enterprise."

[E4] Sphere Fluidics – Cyto-Mine “TTP and Sphere Fluidics introduce Cyto-Mine Single Cell Analysis System” 18.10.2017.

[E5] Synbiobeta – Frost & Sullivan 2017 European New Product Innovation Award 02.11.2017. “Frost & Sullivan Commends Sphere Fluidics for Its Innovative Single-cell Analysis Platform Solution”

[E6] The Scientist – 2018 Innovation Award 01.12.2018. “2018 Top 10 Innovations”

[E7] BusinessWeekly – Geneva Biotech 27.11.2017. “Sphere Fluidics on a Swiss roll with €1.6m for genome engineering project”

[E8] European Pharmaceutical Manufacturer – Oxford Genetics 21.02.2019. “Multi-partner project to automate microfluidic systems for gene editing”

[E9] Sphere Fluidics – ESI-Mine. “ESI-MineTM: high-throughput, miniaturised mass spectrometry using picodroplets”

[E10] Global Health & Pharma – 2018 Biotechnology Awards 23.03.2018. “Sphere Fluidics Limited Most Innovative Single Cell Analysis Platform: ESI-Mine”

[E11] Sphere Fluidics – Products Brochure. “Specialist Research Chemicals & Microfluidic Biochips”

[E12] Sphere Fluidics – Distributors 31.07.2019