Institution: University of Oxford		
Unit of Assessment: 5 – Biological Sciences		
Title of case study: Oxford Biodynamics: EpiSwitch™ chromosomal biomarkers for genomic stratification and personalised medicine		
Period when the underpinning research was undertaken: 2001 – 2015		
Details of staff conducting the underpinning research from the submitting unit:		
Name(s):	**Role(s) (e.g. job title):**	**Period(s) employed by submitting HEI:**
Jane Mellor	Professor	1995 - present
Alexandre Akoulitchev	Research Fellow	2000 - 2007
Aroul Ramadass	Postdoctoral fellow	2004 - 2007
Period when the claimed impact occurred: August 2013 – December 2020		
Is this case study continued from a case study submitted in 2014? N		

1. Summary of the impact

Research into Chromosome Conformation Signatures (CCS) at the University of Oxford led to the spin-out of Oxford BioDynamics to exploit CCS and develop novel biomarkers for diseases including cancer, and immune and neurodegenerative conditions. Their EpiSwitch™ technology is a revolutionary predictive, prognostic and diagnostic microarray platform that is currently being utilised by several companies in experimental and clinical research across a broad range of diseases. The company was floated on AIM in December 2016 and the company's scientific and clinical advances have led to a market capitalisation of more than GBP75,000,000. Their technology is being employed in trials run or sponsored by a range of pharmaceutical companies. Oxford BioDynamics employed 39 staff in 2020, and received the Queen's Award for Enterprise in 2019.

2. Underpinning research

Chromosome conformation signatures (CCS) are higher order structures of chromatin that were originally discovered in yeast as gene loops, through research in the University of Oxford Department of Biochemistry and Dunn School of Pathology [1,2].

Gene loops were first described in 2003 by Mellor and her research group, who demonstrated that Rna15, a factor involved in the 3' end formation of transcripts, was also found at the 5' region of genes [1]. Mellor used this concept to demonstrate that gene loops connected a range of regulatory elements in yeast genes, bringing the promoter and terminator in close proximity in association with RNA polymerase II [2]. This work provided a framework to understand how genes are transcribed, with the terminator being defined before transcription begins. The Mellor group then defined the molecular basis of gene loops and their relationship to non-coding transcription [3].

At this time, Akoulitchev was working with the dihydrofolate reductase (DHFR) gene in mammalian cells and showed how RNA transcripts switched during the cell cycle from non-coding to coding [4]. Using a machine-learning algorithm based on yeast gene loops, and the beginning and end of the DHFR transcripts, the algorithm detected a shared region, which was not a specific DNA sequence but a low free-energy folding region. Putting their diverse observations together, Mellor and Akoulitchev had the idea that these gene loops, more generally described as CCS regions, might mark the potential of a region to be involved in forming higher-order chromatin structure.

Akoulitchev realised that the CCS at the DHFR locus changes during the cell cycle and that this change can be used as a marker of cancer [4]. Based on this insight, the first altered CCSs were defined for cancer (breast and prostate), allowing a test to be developed. The test has proven particularly successful to predict the type of disease a patient has and how they will respond to treatments, especially relevant to the recent revolution in cancer immunotherapy.

Mellor and Akoulitchev were granted an international patent in 2009 that described the measurements of CCS changes in a wide range of applications in disease [5].

3. References to the research

(University of Oxford researchers in bold, students in italics)

Journal articles:

1. **Morillon A, Karabetsou N, O'Sullivan J, Kent N, Proudfoot N,** and **Mellor J** (2003). Isw1 chromatin remodeling ATPase coordinates transcription elongation and termination by RNA polymerase II. *Cell* 115(4):425-35. DOI: 10.1016/s0092-8674(03)00880-8 130 Citations, (WoS 04-2020)

2. **O'Sullivan JM,** *Tan-Wong SM,* **Morillon A, Lee B,** Coles J, **Mellor J,** and **Proudfoot NJ** (2004). Gene loops juxtapose promoters and terminators in yeast. *Nature Genetics* 36(9):1014-8. DOI: 10.1038/ng1411 252 Citations (WoS 04-2020)

3. *Murray SC, Serra Barros A,* **Brown DA, Dudek P,** *Ayling J,* and **Mellor J** (2012). A pre-initiation complex at the 3'-end of genes drives antisense transcription independent of divergent sense transcription. *Nucleic Acids Research.* 40(6):2432-44. DOI: 10.1093/nar/gkr1121. 43 Citations (WoS 04-2020)

4. **Martianov I, Ramadass A,** *Serra Barros A, Chow N,* and **Akoulitchev A** (2007). Repression of the human dihydrofolate reductase gene by a non-coding interfering transcript. *Nature* 445:666-70. DOI: 10.1038/nature05519 479 Citations (WoS 04-2020)

Patent:

5. International patent WO 2009/147386 A1 (2009) Methods for detecting long range interactions in chromatin: **Ramadass AS, Akoulitchev A** and **Mellor EJ**. https://patentscope.wipo.int/search/en/detail.jsf?docId=WO2009147386

Funding for this research included:

Wellcome Trust Programme Grants
N. Proudfoot, 'Transcriptional termination by RNA polymerase II: mechanism and functional role', GBP1,203,275 (062329/Z/00/Z, 2001-2007);
J. Mellor, 'Gene regulation within chromosomal domains: interplay between transcription factors, histone tail modifications and chromatin remodelling ATPases'. GBP639,845 (074557/Z/04/Z, 2004-2009).
J. Mellor, 'Signalling to chromatin: Balancing growth versus longevity', GBP976,045 (089156/Z/09/Z, 2009-2015).

Biological and Biotechnology Science Research Council
J. Mellor, 'Regulation of nucleosome sliding by ISWI chromatin remodelling complexes in vitro', GBP242,876 (43/G17914, 2003-2006)

Human Frontier Science Programme
A. Morrillon, Long-Term Fellowship 'Cell cycle regulation of the CLB2 promoter by the ISWI chromatin remodelling complexes in yeast' (2002)

4. Details of the impact

Pathway to Impact

Oxford Biodynamics was spun out of the University of Oxford in 2007, based on the intellectual property around the CCS [5], to take this technology to market. Much of the early years of the company involved optimising and making reproducible the steps of the CCS measurement process, which was known to be unreliable and thus not yet suitable as a diagnostic tool. Further optimisation resulted in moving from analysis of cancer biopsies to using whole blood. The restriction enzymes, ligase and buffers were optimised and standardised, and all the steps were converted onto a robotic platform. This resulted in a platform that could detect CCS in 4 hours and conversion from a CCS test at individual loci to a microarray platform with 1,200,000 anchoring sites, representing potential CCS throughout the genome. This revolutionised the technology, since many more interactions could be detected. The resulting CCS technology was trademarked as EpiSwitch™. Akoulitchev became the CSO of Oxford Biodynamics, Ramadass the CTO, while Mellor contributed as a member of the company's Scientific Advisory Panel. The EpiSwitch™ platform has worldwide patents, enabling technology licensing across Southeast Asia, Australia, Europe and the United States.

Economic Impact and recognition

In July 2015, OBD raised in excess of USD7,000,000 to accelerate the commercialisation of its epigenetic platform technology – EpiSwitch™; in December 2016 the company was floated on AIM (a sub-market of the London Stock Exchange), and raised GBP20,000,000 from investors. In Aug 2018, GL Capital Group became a 5% investor with approximately GBP9,750,000 new shares in the Company; notably this was GL's first investment in Europe. In December 2019, OBD signed a master services agreement with a major US pharmaceutical company [A], who cannot be identified for commercial reasons.

The patents, licencing agreements and contracts with leading pharmaceutical companies, gave OBD a strong strategic position in the global market and led to a market capitalisation at September 2020 of more than GBP75,000,000 [B]. Revenue in the year ending September 2019 was GBP907,000 [A]. OBD has grown to 39 employees (November 2020, headcount) at sites in Oxford UK (32) and in the USA (3) and Malaysia (4) [C].

OBD won the 2015 Frost&Sullivan European Award for Technology Innovation [D(i)], being described as "the only company with a patented and established biomarker discovery platform for industrial use". In 2019 it received a Queen's Award for Enterprise in Innovation, denoting 'outstanding achievement' and citing the *"Novel epigenetic biomarker technology to deliver personalised medicine*" [D(ii)].

For its validated results with the EpiSwitch™ platform, OBD is recognized and represented in USA at the Foundation of NIH (FNIH) Biomarker Steering Committees in Oncology, Inflammation & Immunity and Neurosciences. [D(iii)].

Benefits to pharmaceutical companies in developing personalised medicine

A number of projects initiated and funded by pharmaceutical companies use the EpiSwitch technology to develop biomarkers that will predict the likely success of a treatment, thus paving the way to personalised medicine.

In a commercial project funded by Merck KGaA, OBD worked with EMD Serono (the biopharmaceutical business of Merck KGaA, Darmstadt, Germany, in the US and Canada), the Mayo Clinic and Pfizer. 99 patients were profiled using EpiSwitch for CCS detection in **non-small cell lung cancer** (NSCLC) using samples and data from the JAVELIN Solid Tumour trial (NCT01772004), to develop and validate baseline predictive biomarkers for response to avelumab [E(i)]. By adding Lyell Immunopharma to the collaboration, they examined biomarkers for response to immune-checkpoint treatments, with samples from patients with NSCLS and **melanoma** [E(ii)]. The CCS approach could identify systemic cellular network deregulations associated with differences in clinical outcome. Furthermore, stratification of patients using CCS outperformed the FDA-approved immunohistochemistry test for PD-L1 expression level, as well as the genetic test based on tumour mutational burden [E(ii)].

Genentech funded the development of the first successful blood-based assay for prognostic stratification and disease subtyping in **diffuse large B-cell lymphoma** (DLBCL), using EpiSwitch to detect, screen and monitor the changes in CCS, in collaboration with Roche and OBD. This work drew on highly-characterized samples and data from a clinical trial funded by Hoffmann-La Roche (NCT00486759) on chemotherapy for DLBCL patients [F]. The results showed 100% effectiveness of the EpiSwitch for CCS detection in classifying DLBCL sub-types from a single blood sample, compared with the extensive tissue biopsy gene expression profiles undertaken in the original trial.

Amyotrophic lateral sclerosis (ALS) is a progressive and ultimately fatal neurodegenerative disease, where motor neurons are lost from the central nervous system, with no known cure. In 2015, OBD won funding from Innovate UK, in collaboration with the University of Oxford and Chronos Therapeutics Ltd, to develop prognostic epigenetic biomarkers for ALS. This enabled ALS diagnosis to be predicted with 75% specificity, with prognosis of fast- or slow-progressing ALS determined to 80% specificity. When the results were published in 2018 [G(i)], a commentary in EBioMedicine the following month stated that the work offered "*... an attractive tool to detect structural related epigenetic changes in ALS*" and posed the CCS as the "*new kid on the block*" for ALS biomarker research [G(ii)].

Subsequently, Mitsubishi Tanabe Pharma America (MTPA) sponsored a study of biomarkers, led by the Massachusetts General Hospital, REFINE-ALS (NCT04259255), in which patient biomarker data from EpiSwitch is compared with assessment of ALS disease progression. The Senior Director of Medical Affairs, MTPA said: "*Through this biomarker study we are seeking to enhance our understanding of edaravone therapy in ALS. We are proud to announce Oxford BioDynamics has joined us in this effort….*" [H]. The first patient was recruited in October 2019 [A] and the study is ongoing.

Patient stratification for COVID-19 severity

EpiSwitch is being deployed in the GETAFIX clinical study funded by the Chief Scientist Office of Scotland to provide epigenetic data to help identify COVID-19 patients at risk of severe disease and to profile patients who will benefit from therapeutic anti-viral treatment with favipiravir. OBD is not a co-investigator on the trial, which is carried out by NHS Scotland and the University of Glasgow [I(i)]. A Professor of Translational Immunology at the University of Glasgow explained the significance of the technology for the study:

> "*To have the greatest impact on the current clinical challenges associated with COVID-19, it is essential that we have the ability to rapidly stratify individuals into those that will progress to severe disease, and those that will respond to available therapies. Evaluation of the immunological set-point via EpiSwitch™ will provide that much needed stratification tool.*" [I(ii)]

By November 2020, OBD was actively engaged in a programme to develop prognostic tests of severity of **COVID-19**, which included work with multiple cohorts from UK, USA, and Latin America, for a total of over 500 patients worldwide [C]. To this end, OBD had secured agreements including a partnership with Boca Biolistics Inc., FL, USA [C,J(i)]; a collaboration with Oregon Health and Science University, Portland, OR, USA (J(ii); and participation in the PREDICT COVID UK Programme (West Hertfordshire NHS Trust) [C].

Training in epigenetics and entrepreneurship

In September 2018, OBD became an industrial partner in a Horizon 2020 Innovative Training Network, 'Predictive Epigenetics' (PEP-NET), which was granted EUR4,000,000 in total. Akoulitchev contributed to the 2020 virtual Summer School, training early-stage researchers in entrepreneurship. Akoulitchev is also the second supervisor for one of the 15 training projects funded by the Network.

5. Sources to corroborate the impact

A. Annual Report and Accounts to 30 September 2020, Oxford BioDynamics. https://www.oxfordbiodynamics.com/wp-content/uploads/2021/03/OXFORDBIO_AR21.pdf

B. Share price of GBP84.50 on 22 December 2020 as shown by London Stock Exchange. https://www.londonstockexchange.com/exchange/prices-and-markets/stocks/summary/company-summary/GB00BD5H8572GBGBXAMSM.html

C. Letter from Chief Scientific Officer, Oxford BioDynamics, 27 November 2020, who is also Corroborator 1, for verification of the numbers and locations of employees.

D. (i) Press release describing Frost&Sullivan European Innovation Award, 22 March 2016. https://www.prnewswire.co.uk/news-releases/frost--sullivan-commends-oxford-biodynamics-for-developing-a-novel-biomarker-discovery-platform-episwitch-573052701.html
(ii) Announcements of Queen's Awards for Enterprise, *The Gazette*, 62621 Supplement 1, 23 April 2019. https://www.thegazette.co.uk/London/issue/62621/supplement/S1
(iii) Membership list of FNIH Biomarker Steering Committees, https://fnih.org/what-we-do/biomarkers-consortium/about/steering-committee

E. Posters at 34th Meeting of the Society for Immunotherapy of Cancer (2019): Shah, EK, Hunter E et al. (i) 'Development and validation of baseline predictive biomarkers for response to avelumab in second-line (2L) non-small cell lung cancer (NSCLC) using EpiSwitch™ epigenetic profiling', P142; (ii) 'Development and validation of baseline predictive biomarkers for response to immuno-checkpoint treatments in the context of multi-line and multi-therapy cohorts using EpiSwitch™ epigenetic profiling', P143. Abstracts available in *Journal for ImmunoTherapy of Cancer* 7(Suppl 1):282. DOI 10.1186/s40425-019-0763-1.

F. Journal article: Hunter E et al (2020). Comparative molecular cell-of-origin classification of diffuse large B-cell lymphoma based on liquid and tissue biopsies. *Transl Med Commun* **5:** 5 DOI: 10.1186/s41231-020-00054-1.

G. (i) Journal article: Salter M et al (June 2018). Initial Identification of a Blood-Based Chromosome Conformation Signature for Aiding in the Diagnosis of Amyotrophic Lateral Sclerosis. EBioMedicine. 33:169-184. DOI: 10.1016/j.ebiom.2018.06.015
(ii) Commentary, Poesen K (July 2018), 'The Chromosomal Conformation Signature: A New Kid on the Block in ALS Biomarker Research?'. *EBioMedicine* 33:6-7. DOI: 10.1016/j.ebiom.2018.07.003

H. Announcement from Oxford BioDynamics plc, 3 May 2019, 'OBD joins ALS Biomarker study sponsored by MTPA' https://www.londonstockexchange.com/news-article/OBD/obd-joins-als-biomarker-study-sponsored-by-mtpa/14062939

I. (i) Glasgow Early Treatment Arm Favirpiravir (GETAFIX) trial protocol: 10.1186/s13063-020-04891-1 (ii) Announcement from Oxford BioDynamics, 30 April 2020, Biomarker platform COVID-19 update https://www.londonstockexchange.com/news-article/OBD/biomarker-platform-covid-19-update/14522642

J. Announcements from Oxford BioDynamics, (i) Strategic partnership with Boca Biolistics, 2 November 2020, https://www.londonstockexchange.com/news-article/OBD/strategic-partnership-with-boca-biolistics/14739535
(ii) Disease severity program for COVID-19 advances, 28 October 2020, https://www.londonstockexchange.com/news-article/OBD/disease-severity-program-for-covid-19-advances/14734540;