**Institution:** Cardiff University

**Unit of Assessment:** Biological Sciences (5)

**Title of case study:** A spinout company for production of organoids at scale

**Period when the underpinning research was undertaken:** 2009-2020

**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: |
|---|---|---|
| Trevor Dale | Professor | 01/09/2003 – present |
| Thierry Jardé | Research Associate | 01/01/2009-31/12/2011 |
| Andrew Hollins | Research Associate | 01/10/2012-31/03/2017, 01/07/2018-26/08/2019 |
| Kenneth Ewan | Research Associate | 01/01/2004-07/10/2020 |
| Elizabeth Fraser | Research Fellow | 01/10/2003-30/06/2017 |
| Mairian Thomas | Research Associate | 01/11/2015-31/03/2016 |

**Period when the claimed impact occurred:** 2013-2020

**Is this case study continued from a case study submitted in 2014?** No

## 1. Summary of the impact (indicative maximum 100 words)

Tumour organoids offer a powerful pre-clinical discovery tool for new anti-cancer drugs. Barriers to commercialisation include culturing organoids with sufficient uniformity and scale to meet rigorous drug development pipeline requirements. Cardiff research developed a novel method for the culture of large quantities of uniform organoids, validated for physiological characteristics. Subsequent collaboration with the University of Bath resulted in a patented bioprocessing technology to grow uniform, reproducible organoids at scale. This research led to a new biotechnology growth-phase company Cellesce Ltd (established 2015), currently valued at £15M. To date, Cellesce Ltd has raised £6M in equity and grants, additionally securing key contracts with multiple customers, including global pharmaceutical and research development companies (e.g., GSK, Merck Millipore and Horizon).

## 2. Underpinning research (indicative maximum 500 words)

Tumour organoids are 3D, self-assembling, multi-tissue cultures that can be grown from patient biopsies. As a miniature version of a patient's tumour, they provide a better predictor of new cancer drug efficacy than traditional 2D cell culture systems. Organoids also offer benefits over 3D spheroid cultures by exhibiting self-assembly and multiple cell types, replicating aspects of the original tumour and its interactions with drugs. Organoids had not previously been grown stably, reproducibly, and on a large enough scale for wider commercial use, despite considerable industry interest in their use for *in vitro* drug discovery pipelines.

The Cardiff team, led by Dale, conducted a programme of research to address this issue, starting with understanding key signalling pathways in mouse organoid models, and continuing through to application in human tumour models and expansion of organoid generation at scale.

### 2.1 Developing and validating organoid models of cancer

#### a. Organoid models of colorectal cancer

The Cardiff team originally began working with organoids to study the WNT signalling pathway **[3.1]**. More than 90% of human colon cancers have mutations leading to the constitutive activation of the WNT/beta-catenin signalling pathway, which normally functions to maintain the unique crypt and villus structure of the intestinal epithelium. WNT signalling is also crucial to the formation of intestinal organoids in culture and is the focus of the Cardiff team's expertise.

In 2012, the Cardiff team established the culture of 3D crypt organoids derived from the small intestine of the Tet-O ΔN89β-catenin mouse. They demonstrated the organoids' utility as an *in vitro* model for the therapeutic targeting of WNT signalling in tumours; specifically, they

showed similar early-stage modulation of gene expression by small molecule WNT inhibitors in organoids compared to equivalent *in vivo* models **[3.1]**.

**b. Broadening applicability of cancer organoids: from colorectal to mammary models**

Funded by Breast Cancer Now **[G3.1]**, the Cardiff researchers further developed the culture conditions for a mouse mammary tissue organoid model. This culture system was extended to two different mouse mammary tumour organoid systems (MMTV-WNT-1 and p53-/+) designed to assay anti-breast cancer compounds and WNT inhibitors (2013–2016) **[3.2]**. This work demonstrated that the Cardiff team's organoid technology could be expanded beyond colorectal cancers, providing important translatability for research on other cancers.

**c. Translation to human cancers and drug discovery**

The applicability of the Cardiff mouse organoid system as a platform for pre-clinical cancer drug testing was further verified by the Cardiff team in a collaborative project with the Merck Group (previously Merck Serono) **[G3.2]**. The Cardiff team showed that the gene expression signature from Tet-O ΔN89β-catenin crypt organoids, treated with a Merck-owned small-molecule WNT inhibitor, mirrored that of data from the mouse model from which they had been derived **[3.3]**.

In further research funded by the existing Merck collaboration, and Cancer Research Wales **[G3.3]** and Cancer Research UK **[G3.4]**, the Cardiff team cultured patient-derived 3D colorectal cancer organoids. They carried out a range of *in vitro* assays using these organoids, which established the benefit of compounds known as tankyrase inhibitors to modify aberrant WNT signalling found in colorectal cancer. These assays established that the inhibitors modulated the expression of genes in the WNT pathway, which the group predicted would reduce the tumorigenic properties of the organoids. When small tankyrase inhibitor-treated cancer organoids were xenografted into immune-deficient mice, the organoids had a reduced capacity to develop into tumours, validating the *in vitro* findings in an *in vivo* model system, and demonstrating expansion of the method to human cancer organoids **[3.4]**.

**2.2 Scale expansion of uniform organoids**

The key issue in applying organoids to high throughput drug screening is the problem of uniformity in size, reproducibility and scale **[3.5]**. While organoids can be directly derived from healthy or diseased patient tissue, they are often heterogenous in nature, and limited in number. To address this bottleneck, the Cardiff team developed a culture method for use in conjunction with bioreactor culturing techniques. The method was designed to facilitate production of large quantities of uniform organoids, which crucially are never grown in contact with plastic (known to alter cell behaviour and reduce *in vitro* assay validity). The team combined their organoid culturing method, with the bioreactor expertise of collaborators from the University of Bath (a team led by Ellis and Argyle). This collaboration led to the development of a bioprocess to culture large quantities of uniform organoids, which the researchers subsequently patented (PCT/GB2017/052026) **[3.6]**. The patented method involved:

1. disaggregating primary organoids to create a single-cell suspension;
2. seeding this cell suspension in a matrix-based culture medium, and culturing in a bioreactor for 24-72 hours, resulting in up to 1 million organoids of varying sizes;
3. recovering these organoids from the matrix medium to create an organoid suspension;
4. sequentially passing the organoid suspension through two sets of cell strainers to exclude organoids above and below specific diameters.

This patented process generates a large quantity of uniform organoids which can be cultured further in the bioreactor, or frozen for storage or shipping. The research subsequently underpinned establishment of a spin-out company (Section 4), which was further supported by proprietary tumour cell lines developed by the Cardiff team **[3.4]**, and a purpose-built bioreactor to support organoid culture, developed by the Bath team.

**3. References to the research** (indicative maximum of six references)

**[3.1] Jardé T**, Evans RJ, McQuillan K, Parry L, Feng GJ, Alvares B, Clarke A and **Dale T** (2013). In vivo and in vitro models for the therapeutic targeting of Wnt signaling using a Tet-O ΔN89β- catenin system. *Oncogene* 32(7):883-93. DOI: 10.1038/onc.2012.103

**[3.2] Jardé T**, Lloyd-Lewis B, **Thomas M**, Kendrick H, Melchor L, Bougaret L, Watson PD, **Ewan K**, Smalley MJ. and **Dale T** (2016). Wnt and Neuregulin1/ErbB signalling extends 3D culture of hormone responsive mammary organoids *Nat Commun* 7:13207. DOI: 10.1038/ncomms13207

**[3.3] Dale T**, et al. (2015). A selective chemical probe for exploring the role of CDK8 and CDK19 in human disease. *Nat Chem Biol* 11(12):973–980. DOI: 10.1038/nchembio.1952

**[3.4]** Badder LM, **Hollins AJ**, Herpers B, Yan K, **Ewan K**, **Thomas M**, et al. (2020). 3D imaging of colorectal cancer organoids identifies responses to Tankyrase inhibitors. *PLoS ONE*. 15(8): e0235319. DOI: 10.1371/journal.pone.0235319

**[3.5] Hollins AJ** and Parry L (2016). Long-term culture of intestinal cell progenitors: an overview of their development, application, and associated technologies. *Curr Pathobiol Rep* 4(4):209-219. DOI: 10.1007/s40139-016-0119-1

**[3.6]** International patent application (number PCT/GB2017/052026, with a filing date of 11th July 2017. The patent application was published on 18th January 2018; reference W02018/011558)

**Selected grants:**

**[G3.1] Dale T** and Clarke A, Defining the role of Wnt signalling in mammary stem cells, Breast Cancer Now, 01/01/2009 – 31/12/2011, £198,643. Funder ref: 2008MayPR16

**[G3.2] Dale T**, Wnt pathway, Merck KGaA, 23/04/2013 – 31/12/2020, £423,449; and **Dale T** and **Fraser E**, Identification of Wnt pathway inhibitors, Merck KGaA, 23/04/2009 -31/07/2013, £1,612,530. Funder ref: CU RCBM776/1

**[G3.3] Dale T** and Clarke A, Developing stem cell containing organoids from primary and metastatic human colorectal cancer for preclinical studies of stratified colorectal cancer therapeutics, Cancer Research Wales, 01/10/2013 – 30/09/2016, £104,280

**[G3.4] Dale T**, Modelling colorectal cancer: Assessing pathway synergies and antagonisms in vivo and validating therapeutic targets, Cancer Research UK, 01/07/2013 – 30/04/2019, £1,682,527. Funder ref. C1295/A15937

**4. Details of the impact** (indicative maximum 750 words)

Based on their novel organoid research, and patented bioprocess, the Cardiff team, together with their Bath collaborators, established spin-out company Cellesce Ltd in 2015. This has now matured from start-up to growth phase, with an estimated value of £15M, and new global biotechnology and pharmaceutical customers and partners.

**4.1 Development of a growth-phase spin out biotech company**

Cellesce Ltd was originally established as BBF BIO LIMITED in 2013. It lay dormant until 2015, when the company name was changed to Cellesce Ltd, and it became the vehicle to commercialise the joint Cardiff / Bath IP and organoid production process. This was based on a business plan developed by the Cardiff team, with Cardiff researcher Dale acting as the company's Director of Organoid Biology since 2016 **[5.1]**.

While some companies (e.g., Stemcell and Merck) have developed products in the organoid market, ranging from specialist growth media to organoids themselves, Cellesce Ltd was one of the first biotechnology companies to focus on the expansion of stable and reproducible human derived organoids at scale. While other manual methods of organoid culture systems support around 1000 assays per batch, the unique patented bioprocessing described in **[3.6]** allows Cellesce Ltd to offer 20-30,000 assays per batch, importantly containing uniform-sized organoids. The organoids available through the company are also validated for morphology,

size range, viability, drug responses and genetics using techniques developed in the Cardiff lab **[3.3, 3.4]**. Further, [text redacted] **[5.2]**.

**4.2 Cellesce Ltd's value and key partnerships diversifying market opportunities**

The global organoid drug discovery screening market was valued at $1.45BN in 2020 (with colorectal cancer worth $136M and breast cancer $500M, *source: Evolution Biosciences*). Cellesce Ltd, operating from its base at Cardiff University's Medicentre (a key University Innovation Hub for clinical and biomedical start-ups), is currently valued at £15M **[5.3]**. The company reported net assets of £720,841 for the 2019/2020 financial year, a twelve-fold increase on the £57,486 net assets reported in 2015/2016 **[5.3]**. The company has just finished a bridging round of fundraising, with a total of £6M raised in equity, convertible loan notes and grants. The company now employs 11 people (eight full-time, three part-time) **[5.4]**.

In 2018, Cellesce Ltd formed a partnership with Repositive, a cancer model platform for researchers in all sectors which allows them to select relevant cancer model systems **[5.5]**. Repositive maintains an inventory of more than 8,000 cancer models and 24 contract research organisations (CROs). The platform matches researchers with the most appropriate cancer model/CRO, helping accelerate their research. Partnership with Repositive means that Cellesce Ltd products and services are visible to Repositive's global customer base, almost half of which are in the US. 85% of Repositive's customers are biopharmaceutical or pharmaceutical companies **[5.5**, p.6**]**. Dr Fiona Nielsen, Co-founder and CEO of Repositive explained that the collaboration with Cellesce Ltd is "*making it easier for oncology researchers and data providers to collaborate and speed up the development of new treatments*" **[5.5]**.

In addition, Cellesce partnered with the Hubrecht Organoid Technology (HUB) which hosts a living organoid biobank of more than 1,000 organoids derived from a variety of organs and disease models. Cellesce's organoid expansion bioprocess is sufficiently flexible that it can be applied to any organoid. Via this partnership, Cellesce's clients and other HUB licensees are scaling production of HUB organoids via Cellesce's patented process **[5.6]**. This partnership provides significant expansion opportunities for Cellesce, as well as providing benefits for HUB. For example, HUB's Managing Director Dr Rob Vries explained: "*HUB organoid technology will benefit from Cellesce's innovative technology to expand large quantities of organoids such as breast cancer organoids*" **[5.6]**.

**4.3 Cellesce's customer base**

By the end of December 2020, Cellesce Ltd had secured contracts with multiple customers, including pharmaceutical companies (e.g., GSK, Merck Millipore), biotechnology companies (e.g., Emulate Inc) and contract research organisations (e.g., Horizon), with initial revenues of £325K secured at the end of 2019 **[5.4]**. For example, Horizon Discovery Group plc is one of Cellesce's commercial partners with an international customer base of over 2,000 organisations, including major pharmaceutical companies, and a revenue of £58.3M in 2019. Horizon is using Cellesce's organoids for drug discovery assays **[5.7]**; Dr Tim Scales, Assay Services Manager at Horizon explained that the company is "*developing screens using organoids as…they offer the next-generation 3D model…which we believe suggests they might offer a better predictive outcome in the clinic*" **[5.8]**. Scales confirmed the benefits of Cellesce's technology: "*Cellesce have a proprietary bioreactor technology that allows them to produce organoids in far greater bulk than normal*" **[5.8]**. He further explained why Horizon selected Cellesce to provide organoids: "*Cellesce's technology allows us to overcome the problems of scalability that we require for high throughput screening but also variation between small batches"* **[5.8]**.

In summary, the Cardiff team applied their research expertise in cancer organoid models to develop a patented bioprocess that forms the intellectual property base of the joint Cardiff-Bath spin-out company, Cellesce Ltd. Since its establishment in 2015, Cellesce Ltd progressed from a start-up to a growth-phase spin-out with an estimated value of £15M, supported by new partnerships (e.g., Repositive) and customers (e.g., GSK and Horizon), who, in turn, command international markets in drug discovery and assay development.

**5. Sources to corroborate the impact** (indicative maximum of 10 references)

**[5.1]** Cellesce webpages confirming Dale as Director of Organoid Biology

**[5.2]** [Text redacted]

**[5.3]** Cellesce Limited Filleted Accounts for five years (2015 – 2020)

**[5.4]** Paul Jenkins, CEO of Cellesce Ltd can corroborate this information

**[5.5]** Repositive webpages and news article detailing Cellesce's partnership with Repositive

**[5.6]** News item detailing the Cellesce's partnership with Hubrecht Organoid Technology HUB

**[5.7]** Horizon web page, application note, and annual report & accounts 2019

**[5.8]** Horizon/Promega video interview with Dr Tim Scales, Assay Services Manager