

EDITORIAL

Organoids: a Special Issue

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I am delighted to introduce this Special Issue of Development, focussing on the emerging field of organoid biology. An organoid can be defined as an artificially grown mass of cells or tissue that resembles an organ. This term has grown in use within the research field over the past few years as a result of advances in two distinct directions: the isolation and propagation of adult stem cell niches in 3D, and the adaption of 3D culture conditions for the directed differentiation of pluripotent stem cell (PSC) lines towards specific developing tissues. Organoids have now been used to model disease, screen drugs, increase our understanding of normal human development and even interrogate biophysical principles of self-organisation. Organoids formed from patient-derived cell types are now in use for the evaluation of drug sensitivities and to validate disease-causing genomic variations. There is no doubt that organoid studies will act as stepping stones to more advanced regenerative medicine approaches, as well as to a better understanding of development, especially human development. Indeed, the field of organoid biology has grown at such a pace, and attracted such attention, that we have devoted this issue of Development to showcase organoid biology. The issue covers the background of the field together with new research studies and technical papers, as well as a review of one of the first scientific meetings devoted exclusively to organoids (Muthuswamy, 2017). In this Editorial, I summarise the common themes that run through the issue and highlight the importance of some of the findings. These articles encompass a broad range of tissue types, model systems and potential applications, and I encourage readers to browse this exciting organoid Special Issue.

Organoids for new tissue types and improvements on previous approaches

The recreation of an adult intestinal niche *in vitro* was a major achievement. Similar adult stem cell-based organoids have now been generated from the epithelium of many adult organs, including other regions of the intestinal tract such as the stomach and colon, as well as the lung and liver. The application of these protocols to disease modelling and the development of personalised drug screening is a major area of growth, and is reviewed here by Drost and Clevers (2017). Although such organoids generally comprise the epithelial niche alone, Jamieson et al. (2017) report in this issue the formation of mammary organoids from the adult mammary epithelium comprising both a secretory epithelium and a surrounding myoepithelial population. A second paper using mammary organoids identifies novel signalling requirements for epithelial stem cell maintenance during development and remodelling (Mohapatra et al., 2017). A non-epithelial population, the human umbilical cord-borne fibroblast, is also reported as capable of creating its own ossicle-bounded, marrow-forming niche

(Pievani et al., 2017). One significant issue in the field is how to scale up organoid production in terms of both organoid size and efficiency. Sachs et al. (2017) describe the coalescence of multiple adult intestinal crypts to generate a more anatomically patterned intestinal segment with villus and crypt-like features. While these do not spontaneously arise from the initially isolated cell populations, they can be formed from a series of classical gut-derived organoids, generating a more anatomically correct tissue at the macroscopic and microscopic levels.

Human PSC-derived organoids offer the opportunity to investigate specifically human developmental biology, as reviewed by McCauley and Wells (2017) in this issue, and thus can bring insight into how to better generate more functionally relevant cell types *in vitro*. With this in mind, Voges et al. (2017) report on the generation of heart organoids and use them to investigate the regenerative capacity of human fetal heart tissue *in vitro*. While the formation of cardiomyocytes has long been possible in 2D, researchers are now tweaking protocols for generating PSC-derived organoids to either improve specification or patterning in 3D, which will have important implications for both drug screening and disease modelling. PSC-derived organoids are also being used to understand normal human development, as exemplified by Tsai et al. (2017), who investigate the signalling mechanisms regulating regional identity in the human intestine. Using PSC-derived cerebral organoids, Matsui et al. (2017) uncover new mechanisms that regulate growth, survival and neuronal migration.

What can organoids tell us about morphogenesis?

The remarkable capacity for cell types to recreate complex multicellular structures in 3D, and indeed to maintain such structural complexity across time and repeated dissociation in some instances, is a testament to the power of morphogenesis and cellular self-organisation. Indeed, organoids can be used to interrogate the driving principles of morphogenesis, and the opportunity for synergy between organoid biology and materials science is boundless. In their Spotlight article, Dahl-Jensen and Grapin-Botton (2017) discuss how the field of physics can help our understanding of organoid formation, while Gartner and colleagues (Murrow et al., 2017) review engineering-based approaches to dissect stem cell-niche interactions. The organoid field arguably grew out of seminal studies in the 1900s based on the capacity for disassociated cells to reform structure. In this issue, Lefevre et al. (2017) return to the original approach of dissociating embryonic tissue to examine the forces that drive self-organisation in the context of the developing kidney. While predicted as long ago as 1955 (Townes and Holtfreter, 1955), this study again revealed a requirement for selective adhesion between like cell types. Eldred et al. (2017) also use self-organising aggregates to investigate the role of cell-cell interactions in driving tissue morphogenesis, this time in the context of neural lamination. Some previous approaches to the generation of organoids have involved the combination of stem cell-derived somatic cell types with a source of endothelium and mesenchymal stem cells. In this way, Takanori Takebe has

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previously developed organ bud models for a variety of tissues, and Asai et al. (2017) have now begun to dissect this system by deconstructing their own liver organoid protocol to specifically ask whether secreted growth factors or direct contact is driving the process. Koike et al. (2017) look at the role of branched-chain amino acid metabolism in the developmental expansion of embryonic liver progenitors, and apply these findings to selectively amplify self-renewing bi-potent hepatic progenitor cells in human liver organoid cultures. Also in this issue, Giacomelli et al. (2017) report the combination of cardiomyocytes and endothelial cells for forming 3D heart microtissues. The significant improvement in morphogenetic patterning observed with the addition of these two cell populations begs the question of their role in morphogenesis. A better understanding of tissue morphogenesis and cell-cell interactions will require, in part, more sophisticated tools for imaging, and here organoid technology is particularly advantageous. In this context, Saarela et al. (2017) report a novel kidney organoid culture and imaging system based on fixed z-dimension (FiZD) time-lapse confocal imaging.

...and what of the challenges?

At present, the hype around what we might ultimately be able to do with what are currently tiny anatomically imperfect structures is rife. As with other advances in science, it will be important moving forward to balance the hype with the reality and hence not fuel hope in inappropriate ways. Hence, the issue opens with a thought-provoking piece (Huch et al., 2017) consisting of two parts: the hope of organoid research written by Huch and Knoblich, and the hype – including what it will take to overcome it – written by Lutolf and Martinez-Arias. Indeed, organoid culture is also not without some ethical challenges, as discussed in the Spotlight article from Munsie, Hyun and Sugarman (Munsie et al., 2017). The generation of human cerebral cortex in the dish, particularly coupled with genome editing, will require ethical consideration. Modelling early human embryological development in 3D, as discussed in a Review by Simunovic and Brivanlou (2017), will also require careful ethical consideration.

At a far more practical level, these new approaches for generating models of complex multicellular tissues bring with them substantial challenges with respect to robustness and reproducibility. For example, the use of patient-derived induced pluripotent stem cells (iPSCs) to model my favourite organ, the kidney, will be substantially challenged by the fact that these organoids contain more than ten distinguishable cell types. Minor variations in the relative ratios of any given cell type between differentiations of even the same cell line are likely. This will be substantially compounded when comparing organoids generated from different clones, let alone distinct individuals. In an effort to improve the robustness of organoid formation, Arora et al. (2017) report a process engineering approach that increases the formation of high-quality intestinal organoids by up to 3.8-fold, based on automated micropipette sorting at the spheroid stage. Perhaps more pertinent is the reality that all iPSC-derived organoids generated to date represent a model of the prenatal state, not a fully differentiated tissue type, let alone one from an aged individual. Whether we can use them, therefore, to predict more than monogenic/oligogenic early-onset developmental disorders must be established tissue by tissue. There is promise, however. Organoids have been meaningfully applied to model tissue responses to viral and bacterial infection, such as the application of cerebral organoids to the study of Zika virus, discussed here by Song, Ming and colleagues (Qian et al., 2017). Lung organoids, reviewed extensively by Hogan and colleagues (Barkauskas et al., 2017),

might also prove highly amenable to modelling viral and bacterial infection, as well as other agents that are known to damage the airways. Whereas modelling tissue-specific diseases has proven informative, models of multi-organ system chronic disease will be less likely to advance using organoids.

It has been a great pleasure bringing together this collection of articles featuring the state of the art in organoid biology. We hope that it provides the reader with both a resource of background information and a snapshot of the latest advances. I have no doubt that there is much more to come. The application of organoid technology to address comparative anatomy and associated evolutionary questions will undoubtedly bring fascinating insights, and our dissection of the primary drivers of organoid morphogenesis will advance regenerative medicine. Although the direction in which this field will move in the coming years is yet to be seen, this is undoubtedly an area in which a return to fundamental cell and developmental understanding has and will continue to play an important role.

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