Developmental Cell

CellPress

Voices

20 years of *Developmental Cell*: Looking forward

In our 20th anniversary year, we reflect on how fields have changed since our first issue and here look to the future. In this collection of Voices, our writers speculate on the future: in terms of philosophy, cell states, cell processes, and then how to model cell systems.

Takashi Hiiragi
Hubrecht Institute, Utrecht, Netherlands

New questions!

It is an exciting time for cell and developmental biologists. Technical innovations are enabling us to see, probe, and measure what was inaccessible just yesterday. These technical developments will continue, and in 10–20 years, we will have essentially no limit in doing and getting what we want. In the meanwhile, there may be a rate-limiting step in analyzing massive amounts of data, but ever-improving computational power and the development of AI will provide solutions. A paradise for us scientists—or is it?

Well, for it to be, we need to know what we want to know. The key role for scientists would be in defining the question.

Collecting comprehensive data, from omics to screens, may become a matter of clicks. Scientists would focus on identifying new questions and designing the study. With so much more visible and open to analysis, we will be able to go in depth in biology, ask bold questions that were previously unapproachable, define long-standing questions in a tractable manner, and discover entirely new questions. While methods will be streamlined, study design will remain dependent on us, individual scientists.

Science will become more personal. It will be the question, idea, and style that identify scientists—like an artist identifiable by their art. Be creative. Science will be once again closer to art.

Amy S. Gladfelter
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Dwelling in the differences

Biomolecular condensates have now been uncovered in almost every corner of the cell. An enormous amount of work remains ahead to determine how the physiochemical features of condensates influence biochemistry. What are the common emergent features of all condensates, and how do the myriad of assemblies differ? It is the differences that are difficult to embrace as we are drawn to name common patterns. However, I think the differences are essential to the vitality of the field.

Just as, as a culture, we are working to expand our language to respect a plurality of identities of people, we also need to consider language in the field of phase separation that is expansive yet precise. There is a need to accept that there are many possible mechanisms for assembly and function that coexist. We need to find meaning from the diversity of forms that arise from the complex array of molecules that use phase separation to gather.

This vision comes with challenges in how we communicate, how we share data and protocols, and how we remain curious and not dogmatic about singular mechanisms when grappling with such a wide array of cellular structures. My dream is that in practicing tolerance and openness in our scientific models, this also shapes the culture in the field to be inclusive to not only many disciplines but also to an array of scientists that reflect the diversity of the world.

Irene Miguel-Aliaga
MRC London Institute of Medical Sciences, Imperial College, London, UK

Adding new dimensions to inter-organ communication

For hundreds of years, scientists have wondered how organs communicate. We are now able to tackle this question with unprecedented granularity. Moving beyond the "textbook" roles of peptide hormones, we are revealing inter-organ exchanges of metabolites, nucleic acids, organelles, and even microbes. I am curious to see what else organs trade and why; I suspect that we have underestimated the importance of mechanical cues in this context.

Advances in interrogating and interfering with gene expression with spatiotemporal resolution will allow us to explore the geometry and dynamics of inter-organ crosstalk. If we manage not to get lost in the details, we might be able to reveal whether there is a 3D spatial logic to how organs are positioned relative to one another and how they communicate. Similarly, by describing information sequences over time, rather than taking snapshots, we should move beyond identifying inter-organ signals to reveal an inter-organ language of sorts; might that reveal some inter-organ grammar?

As we uncover these new layers and dimensions, we will learn about the tripartite interactions between gene regulation, mechanics, and metabolism that make organs grow, remodel, and decline, blurring the somewhat artificial boundary between developmental biology and physiology.

Finally, as we survey communication across scales (from inter-organelle to inter-organ crosstalk), we will not only reveal general features of information transfer but also shed light on the key biophysical constraints that confine certain information exchange features to particular scales.

Hilary A. Coller
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A molecular framework for "chilling"

A yeast far from the grapes that nourish it. A lymphocyte that hasn't encountered its cognate antigen in years. A spore waiting for its time to germinate. All of these cells enter a distinct state teenagers might call "chilling." Cells in this quiescent state of temporary and reversible cell-cycle arrest readjust their metabolic pathways to reduce unneeded biosynthetic processes. They activate protective pathways that reduce the accumulation of unwanted chemical reaction products that degrade the integrity of macromolecules. They sample their environment for cues to proliferate again. Quiescent cells, in many species and contexts, maintain their capacity to create functional progenitors at the appropriate place and time.

While significant progress has been made in understanding many aspects of quiescence, there remain important unanswered questions. What are the critical molecular changes that allow for a reversible quiescent state, and how are these molecules similar or different depending on the quiescence signal, the species, and the tissue? What are the key properties that distinguish quiescent cells from cells that exit the cell cycle more permanently, such as terminally differentiated cells? How is depth of quiescence achieved by longer time or a more stringent stimulus related to the reversibility of quiescence, senescence, and aging? Understanding the molecular basis for these key properties of quiescent cells will shed light on the fundamental rules for how organisms survive in diverse environments, develop, and protect and repair themselves.

On Sun Lau
National University of Singapore, Singapore

Developmental plasticity at cell lineage resolution

A phenotype is the product of genotype and the environment. Modern developmental biology has had tremendous success in revealing genetic programs that specify forms and patterns. A major question that follows then is how environmental factors influence these programs and drive plasticity in phenotypes. How stable will the generation of forms and patterns be in fluctuating environments? What are the environmental factors that impact this? How are extrinsic cues relayed to cells, integrated, and able to alter cell-fate decisions? Addressing plasticity is important to understand how development occurs in the real world, which has direct relevance to medicine and food production.

The development of stomata, the epidermal pores of plants for gas exchange, offers an exciting and accessible gateway to study plasticity of a cell lineage. As stomata directly control plant physiology, their production by the stomatal lineage is receptive to diverse external signals such as temperature, light, and pathogens. A network-level understanding of the various signals that intersect with the intrinsic stomatal pathway would depict how cells incorporate complex signals in making fate decisions. Achieving this will require careful dissection of the environmental pathways in a cell-type-specific manner as well as novel techniques that probe for transcriptomics/epigenomics and interactomes at the single-cell level. Given the parallels between stomatal lineage and adult stem cell lineages in animals, the study of stomata may inform broad principles underlying cell and lineage plasticity.

Heidi M. McBride
Montreal Neurological Institute, McGill University, Montreal, QC, Canada

Keeping up with the maverick mitochondria

I was a postdoc when the first Fzo1 paper came out 24 years ago, and it was instantly clear that a new field was about to launch. Indeed, work in yeast rapidly defined the key molecules and steps required for mitochondrial fusion and fission, clinicians identified mutations in the mammalian orthologs of the yeast proteins causing disease, and the field grew from just a few wayward cell biologists to a multidisciplinary explosion of research into mitochondria as "more than just a powerhouse."

Many questions have been answered about the fundamental processes of mitochondrial dynamics, yet we see surprising new phenomena reported as the maverick mitochondria keep breaking all the rules. Mitochondria can jump ship and spread between cells; mtDNA and mtRNA get released into cytosol to drive inflammation; fission requires not just the ER but also lysosomes and Golgi; actin comet tails scatter mitochondria during mitosis—all concepts I couldn't have imagined. The fact that we keep seeing such unexpected things tells me we've only explored the tip of the mitochondrial iceberg and that the future can only be met with an open mind.

With all new discoveries, it takes time and persistence to develop robust, quantitative assay systems to fully resolve mechanisms, which must be a focus moving forward. The next 20 years will also see a heightened interest in these questions as the field continues to expand across scales to explore mitochondrial contributions to cell-fate decisions, in tissue organization and disease pathologies. It will be an exciting challenge for us all.

Kazuhiro Aoki
National Institutes of Natural Sciences, Okazaki, Japan

ERK dynamics and the directions they will take us

With the advent of genetically encoded fluorescent biosensors and live-cell imaging, cell signaling dynamics has attracted attention as a potential mechanism for cell-fate decisions. The concept of "dynamic coding" has been well characterized with certain cell signaling molecules. Especially, the spatiotemporal dynamics of ERK MAP kinase activation have been visualized in cultured cells and model organisms, demonstrating the role of ERK dynamics in a wide range of cellular functions. However, several challenges still remain to be overcome.

The first challenge is to understand the encoding mechanism; what kind of input stimuli generates distinct ERK dynamics through signaling networks? ERK dynamics often exhibit excitable patterns, but the source of the noise remains largely unknown. Additionally, although intracellular signaling has been the main focus to date, it will also be necessary to visualize the extracellular ligands. The second challenge is to clarify the decoding mechanisms; how are specific ERK dynamics being filtered and interpreted, resulting in distinct cellular phenotypes? As ERK dynamics alone are unlikely

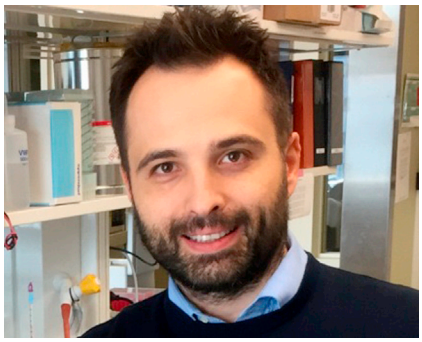
to explain the diversity of cell-fate decisions, it will be necessary to consider the possibility of a combination of other signaling dynamics with ERK to reveal the decoding strategies. The third challenge is to investigate whether there are species-specific ERK dynamics and their evolutionary implications. A promising approach is to visualize the spatial and temporal ERK dynamics *in vivo* in non-model organisms, which have been largely underappreciated.

Andreas Linkermann
Clinic of Internal Medicine III, University Hospital Carl Gustav Carus at the Technische Universität Dresden, Dresden, Germany

The rush on ferroptosis: Into the Iron Twenties

In the Golden Twenties, iron was out. Steel was in, and gold of course! But things change. In 2012, Dixon and Stockwell coined the term "ferroptosis," which has ignited a boost of 2,500 publications. The ongoing gold rush is driven by high hopes. First, control of ferroptosis is fundamental to all existing life. Second, ferroptosis is a driver of stroke, myocardial infarction, acute kidney injury, and many other clinical conditions. Third, persister cells of several cancers depend on anti-ferroptotic systems. Fourth, ferroptosis causes the loss of pancreatic islets upon preparation for transplantation. Finally, the quality of solid organ transplants may benefit from ferrostatins.

Often you will not see the detail in your path if you are rushing at top speed. While clinical trials are initiated, we still do not understand the critical details and consequences of ferroptosis. In fact, we do not even understand the physiological function of ferroptosis in general. While we are aware of damage-associated molecular patterns (DAMPs) that are released from cells that die by ferroptosis, we do not know how an immune response is shaped by ferroptotic debris. We also do not understand how ferroptosis allows regeneration of tissues in some settings while it leads to fibrosis in others. Convincing the FDA to approve ferroptosis-targeting therapies and clinicians to use them requires solid answers. Most importantly, we do not have a biomarker for ferroptosis. But thousands of ferroptosis researchers around the world are keen on finding one. For the sake of our patients, keep searching!



Stefano Santaguida
European Institute of Oncology and University of Milan, Milan, Italy

Aneuploidy and CIN take the stage

Aneuploidy and chromosomal instability (CIN) strongly correlate with aggressive malignancies and poor patient outcomes. Nonetheless, we still lack a comprehensive understanding of their role in tumorigenesis. This reflects the difficulties associated with manipulating single entire chromosomes compared to relatively more easily accessible methods available to study a single gene(s).

An important goal in the near future will be to build and exploit reliable systems *in vitro* and *in vivo* to enable a thorough understanding of the effects of karyotype changes on cell physiology. The coming years will also be the appropriate time to address the role of chromosomal instability in both primary tumors and metastases, through controlled generation of periods of CIN. Fundamental progress will undoubtedly be rooted in careful molecular characterization of the consequences of aneuploidy and CIN in terms of both proteotoxic and genotoxic stress, as well as with regard to the delicate and intricate relationship with inflammation and immune clearance.

All of this will be achieved through detailed molecular analyses *in vitro* coupled with accurate studies *in vivo* and will be fostered by tight collaborations among scientists in the field to integrate different expertise. Exciting times are ahead of us and hold the promise of revealing how aneuploidy and CIN play a role in cancer, with the hope of facilitating the development of new therapeutic interventions targeting the aneuploid state of cancer.

Geoffrey Wasteneys
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The cytoskeleton: A key driver of plant development

Plant cytoskeletal research is immersed in understanding how microtubules and actin filaments orchestrate morphogenesis through specification of division planes, placement of cross walls, and direction of cell expansion. This focus portrays cytoskeletal machines and arrays at the end of a hierarchical chain of command, initiated by environmental cues. In the next decade or two, I predict that we will think of the plant cytoskeleton as not merely a downstream effector but rather as a key sensor of environmental signals and an essential modulator of developmental plasticity. This vision is informed from my own involvement in the discovery of a negative feedback loop involving the microtubule-associated protein CLASP, which both controls the organization of microtubule arrays and modulates brassinosteroid signaling and auxin transport in the root apical meristem.

Future cytoskeletal research will yield conceptual advances in plant evolutionary development, such as the emergence of plant meristems and vascular tissues, organ plasticity, and metabolic responses to abiotic and biotic stress. I predict in the coming years that we will become less reliant on *in vitro* biochemical assays and more adept at quantitative biochemical experiments in living cells, within intact organs subjected to realistic environmental conditions. We will see the integration of quantitative imaging techniques such as FLIM and FRAP with mathematical modeling. Genome editing, recombineering, photoconvertible fluorescent proteins, and optogenetics will also play important roles in this revolution.

Nicolas C. Rivron
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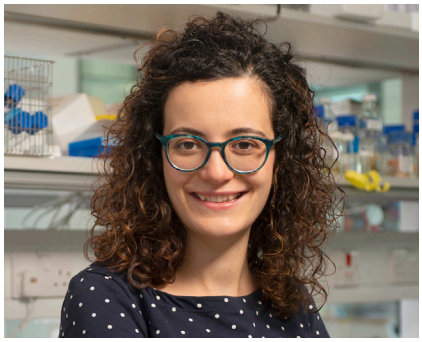
Modeling early development with blastoids

The first weeks of mammalian development are crucial: defects not only cause infertility but also contribute to long-term health problems decades later (e.g., cardiovascular). However, these stages remain less understood due to poor accessibility.

Blastoids are blastocyst models formed by stem cell self-organization. Because they mimic the pre-implantation stage, they can be transferred *in utero* (mice) or combined with endometrial organoids *in vitro* (human) to better understand the fragile processes of implantation and early development. Ongoing efforts aim at assessing how accurate and predictive blastoids are. This will depend on their ability to recap the sequence and timing of specification and morphogenesis, while forming only the right cells.

Current stem cells (mouse and human) reflect the mid-blastocyst stage. In the future, establishing stem cells reflecting earlier stages as well as from other species will allow for modeling earlier embryos and studying evolutionary aspects. Combining blastoids with more complete uterine organoids will also offer ways to reveal *in vitro* how we and other species develop.

Blastoids are models that cannot be used for direct reproduction, and transferring human blastoids *in utero* is forbidden by the ISSCR. They are, however, an ethical alternative for scientific studies and might guide clinical practices to better manage early pregnancy. This is a huge lever to enhance gender equality while supporting the health and progress of communities worldwide through effective family planning and disease prevention.

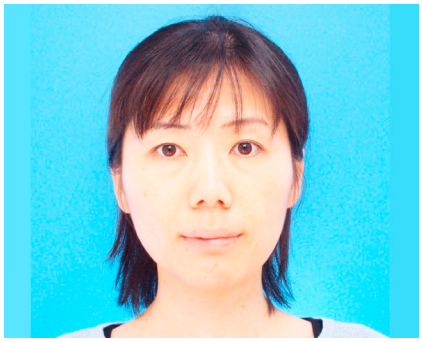


Marta N. Shahbazi
MRC Laboratory of Molecular Biology, Cambridge, UK
Image courtesy of MRC Laboratory of Molecular Biology

Developmental biology meets human reproduction

Why is human reproduction so inefficient? This is a fundamental yet unsolved question. While model organisms offer important insights to early development, the lack of experimental platforms to study human embryogenesis has limited developmental biologists and hindered our mechanistic understanding of human reproduction. However, things have started to change. Assisted reproductive techniques generate vast numbers of embryos that cannot be used for clinical treatment and hence represent a precious resource for research. Novel culture methods, genome editing tools, sequencing, and imaging techniques can now be applied directly to human embryos to unveil the molecular and cellular mechanisms of our development and how they may go awry during pregnancy loss.

Our second essential tool is human stem cells. Under the right conditions, they self-organize and recapitulate morphogenetic and patterning events of human embryogenesis. These models provide flexibility, tractability, the possibility for high-throughput experimentation, and fewer ethical considerations than intact human embryos. They are especially relevant to study gastrulation, the emergence of the body plan, and how alterations in these events lead to miscarriage and congenital malformations. Last but not least, the generation of gamete-like cells from embryonic stem cells allows us to explore the mechanisms of human gametogenesis, a fundamental step toward tackling infertility. Early pregnancy loss, miscarriage, and infertility are concepts that now drive clinical embryologists, gynecologists, and developmental biologists alike.



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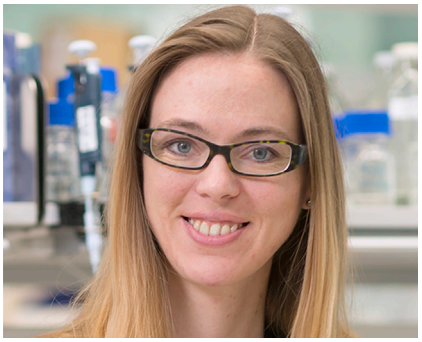
In vitro models of somitogenesis

A trend of somitogenesis research is the use of *in vitro* models. The human and mouse segmentation clocks were recapitulated from pluripotent stem cells, and even somite-like structures were periodically formed in mouse embryonic organoids, gastruloids. Researchers will create more mature organoids that form dermomyotome and sclerotome, or even skeletal muscles and bones. In parallel, efforts will be made to create organoids that form not only somites but also the surrounding tissues. Since pluripotent stem cells of versatile mammalian species are available, creating similar stem-cell-derived models should also be possible for unconventional animals, such as rhinoceros. Although those *in vitro* models do not fully reflect *in vivo* conditions, they are still valuable tools that make imaging, measurement, and manipulation more accessible than living embryos.

Then the question is how to utilize the novel *in vitro* models. Human somitogenesis models will help with understanding congenital diseases of body axis patterning. Comparing multiple species in the same *in vitro* environment will clarify both conserved mechanisms and inter-species differences in somitogenesis (bonus: bovine and porcine muscle derived from somites can be edible cultured meat). New frontiers in somitogenesis, such as tissue mechanics and metabolism, should also benefit from *in vitro* models. Ultimately, researchers will try to manipulate somitogenesis with *in vitro* models. Manipulating the speed of the segmentation clock and the size of somites may pave the way to control the time and size in development.



Madeline A. Lancaster
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Next-generation organoids

Organoids are powerful tools for studying organ development and function in a human context. The organoid field has made remarkable progress over the past decade, and if the past is any predictor, the future is likely to be bright indeed. We will see increasingly complex methods involving bioengineering to improve structure and function, increasing cellular complexity to recapitulate true organ makeup, and even inter-organ interactions through linking organoids of different types. The major challenge of vascularization will continue to be a topic of focus, but if this hurdle can be overcome in a way that truly improves organoid development and function, then the payoff will be huge. Such advanced organoids with ever closer structure and function to the real thing would enable discoveries of cellular and molecular mechanisms unique to humans and even evolutionary studies to identify creative ways nature has found to accomplish difficult biological tasks.

At the same time, simpler organoids with improved reproducibility that model specific aspects of the organ in question will be developed for studies such as high-throughput screening and disease modeling. The challenge here will be maintaining a faithful representation of the organ while optimizing protocols for ease of use and scale-up. But the benefits could lead to new improved therapies and even cell or tissue replacement approaches for patients with compromised organ function. Overall, the organoid field will blossom with increasingly diverse methods for a variety of developmental and cell-biological questions.