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MEETING REVIEW

On growth and force: mechanical forces in development

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ABSTRACT

The EMBO/EMBL Symposium 'Mechanical Forces in Development' was held in Heidelberg, Germany, on 3-6 July 2019. This interdisciplinary symposium brought together an impressive and diverse line-up of speakers seeking to address the origin and role of mechanical forces in development. Emphasising the importance of integrative approaches and theoretical simulations to obtain comprehensive mechanistic insights into complex morphogenetic processes, the meeting provided an ideal platform to discuss the concepts and methods of developmental mechanobiology in an era of fast technical and conceptual progress. Here, we summarise the concepts and findings discussed during the meeting, as well as the agenda it sets for the future of developmental mechanobiology.

KEY WORDS: Mechanical forces, Mechanobiology, Mechanosensation, Mechanotransduction, Morphogenesis

Introduction

How an embryo, initially consisting of a single cell, can grow and shape itself into a complex organism is a question that has been tantalising scientists for more than a century. In his 1917 opus magnum *On Growth and Form* the British polymath Sir D'Arcy Thompson, trying to provide an answer to this question, wrote: 'The form, then, of any portion of matter, whether it be living or dead, and the changes of form that are apparent in its growth, may in all cases alike be described as due to the action of force. In short, the form of an object is a "diagram of forces".' (Thompson, 1917). A little more than a century later, the idea that mechanical forces act in concert at all scales to generate biological shapes remains more relevant than ever. This was vividly illustrated by the interdisciplinary community of scientists who gathered this summer in Heidelberg (Germany) to discuss the role of 'Mechanical Forces in Development' at the EMBO|EMBL Symposium organised by Naama Barkai (Weizmann Institute of Science, Israel), Enrico Coen (John Innes Centre, UK), Carl-Philipp Heisenberg (IST, Austria) and Frank Schnorrer (IBDM, France).

Thanks to the development of quantitative approaches and new techniques such as light-sheet or super-resolution microscopy, single cell 'omics', genome editing techniques, optogenetics, mathematical modelling and deep learning, the field has advanced considerably over the last two decades (Heisenberg and Bellaïche, 2013). These advances have revolutionised how we watch and perturb morphogenetic events in an ever-expanding list of model organisms and *in vitro* systems. Maybe more importantly, and in spite of previously existing disciplinary divides, they have contributed to the field forging itself as a new discipline – 'developmental mechanobiology' – wherein theory and experiment are intimately intertwined.

Despite this progress, a number of questions remain open. How are mechanical forces generated and transmitted across scales? How are they sensed and converted into molecular signals? How are mechanosensors precisely tuned? As we review below, these issues were the focus of talks, poster sessions and intense discussions that took place during the meeting.

Mechanics of cells and tissues

Mechanics in individual cells

Several speakers reported on progress in understanding the mechanobiology of individual cells. The keynote lecture by Ewa Paluch (University College London, UK) summarised her lab's long-standing efforts to determine the molecular composition and fine structure of the actomyosin cortex using proteomics (Biro et al., 2013) and super-resolution microscopy (Clark et al., 2013). She then discussed the effect of cortical tension on cell differentiation, showing that a decrease in tension correlates with, and is required for, the differentiation of embryonic stem cells via ERK signalling (De Belly et al., 2019 preprint). Verena Ruprecht (CRG Barcelona, Spain), building on her earlier study of a confinement-induced amoeboid mode of migration in embryonic zebrafish cells (Ruprecht et al., 2015), reported important advances on the identification of the signalling pathway that controls the switch between different modes of cell motility through nucleus deformation, calcium release and a mechanosensitive nuclear phospholipase (Venturini et al., 2019 preprint). Ulrich Schwarz (Heidelberg University, Germany) discussed an intriguing aspect of malaria infection, whereby *Plasmodium falciparum* orchestrates a 're-engineering' of the red blood cell cytoskeleton, making the cells switch from their canonical biconcave shape to a spherical one, and covering them in thousands of protrusions rich in parasite-generated adhesion molecules. By becoming adhesive to the endothelium, the malaria-infected red blood cells stay for longer in the vasculature and thus avoid clearance by the spleen.

Other talks discussed the mechanics of cells within tissues. Caren Norden (Instituto Gulbenkian De Ciência, Portugal) addressed interkinetic nuclear migration in neuroepithelia. Surprisingly, the underlying cytoskeletal processes differ between straight (hindbrain) and curved (retina) neuroepithelia, involving Rho-ROCK-Myosin in the first case and formin-mediated pushing in the latter (Yanakieva et al., 2019). Thus, even the apparent simplicity of a one-dimensional migration event can hide surprisingly variable mechanisms. Frank Schnorrer (IBDM, Marseille, France) discussed *Drosophila* myofibril formation, illustrating how the recent development of new molecular sensors for mechanical forces in tissues helped unravel the function of talin as a molecular force transmitter (Lemke et al., 2019). Finally, Ozge Ozguc (Institut Curie, Paris, France) discussed emerging insights into an intriguing phenomenon observed in the pre-implantation mouse embryo: the

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existence of periodic, travelling contraction waves across the cortex of blastomeres (Maître et al., 2015).

Mechanics of epithelia: geometry, robustness and deformations

One of the basic building blocks of organisms are epithelial tissues, the properties of which have raised multiple alluring questions for biophysicists (Lecuit et al., 2011). For example, epithelial cells are arranged in characteristic spatial patterns, reminiscent of physical structures such as foams, yet they are able to undergo homeostatic turnover whilst ensuring robustness of the overall tissue architecture. Moreover, their ability to undergo long-term, controlled deformations is fundamental to morphogenesis. Several elegant talks addressed the emerging molecular and biophysical understanding of these phenomena.

It has long been known that epithelia exhibit a viscoelastic behaviour (Petridou and Heisenberg, 2019): as they return to their resting shape in response to a transient force of deformation, they slowly and irreversibly deform by dissipating mechanical stress in response to a more sustained mechanical stimulus. Using a combination of live imaging and optogenetics, Margaret Gardel (University of Chicago, USA) showed how irreversible deformation of epithelia can be generated by pulsatile RhoA activity via pulsatile ratchet-like shortening of intercellular contacts that relies on mechanosensitive E-cadherin endocytosis (Cavanaugh et al., 2019; Staddon et al., 2019). In a complementary talk, Venkatesan Iyer (MPI Dresden, Germany) showed that mechanosensitive E-cadherin endocytosis is controlled by p120-catenin acting as a stress sensor and tunes the viscous properties of the *Drosophila* wing epithelium (Iyer et al., 2019).

Michel Labouesse's team (IBPS, Paris) showed that embryonic antero-posterior elongation in *Caenorhabditis elegans* proceeds in an unusual fashion: forces generated by muscle contraction trigger irreversible deformations of epidermal cells. Combining modelling and molecular experiments, they showed that muscle contractions induce shortening of the actin microfilaments in epidermal cells due to the action of actin-severing proteins. Severed actin filaments are then stabilised by the action of SPC-1 and PAK-1, which act as a molecular lock ensuring the irreversible deformation of the cells, resulting in the embryo deforming as a viscoplastic solid (Lardennois et al., 2019).

Though deformable, epithelia are also robust. For example, Naoto Ueno (NIBB, Okazaki, Japan) presented a proteomic investigation of the effect of compressive stress on early *Xenopus* embryos, revealing that compression triggers phosphorylation of focal adhesion and tight junction components, resulting in tissue strengthening (Hashimoto et al., 2019). Similarly, Beth Pruitt (University of California, Santa Barbara, USA) explained how applying exogenous shear along a line in the middle of an epithelial monolayer results in oscillations propagating across the tissue, which are dampened by actin filament turnover (Sadeghipour et al., 2018).

A long-standing problem is the spatial organisation of cells in epithelia. Marco Kokic (ETH Zürich, Switzerland) discussed Lewis' law, an empirical rule proposed almost a century ago that states that the apical surface area of a cell is proportional to the number of its nearest neighbours. Kokic suggested that Lewis' law emerges through cell surface energy minimisation, resulting at the tissue scale in cells taking the most regular polygonal shapes within a contiguous lattice, thus minimising the average perimeter per cell and, thereby, the contact surface energy between nearest neighbours (Kokic et al., 2019 preprint).

Finally, Adam Ouzeri (LaCàN, Barcelona, Spain) presented a new theoretical model attempting to bridge scales between molecular-level active-gel models of the cell cortex and tissue-level phenomenological vertex models. Such a framework provides a unified representation of tissues during homeostasis and development, allowing the description of some seemingly disconnected mechanical behaviours, such as stress relaxation or creep behaviour upon tissue stretching, tissue buckling upon compression, tissue-scale pulsatile contractions, and active superelasticity.

Tissues under threat: when forces challenge homeostasis

One of the defining features of a homeostatic system is its ability to return to a resting state in the face of external perturbations. Several talks explored how epithelia defend themselves against mechanical challenges – either externally imposed (e.g. injuries) or internally generated (e.g. by over-proliferative cells).

Alpha Yap (University of Queensland, Australia) summarised how epithelial homeostasis is maintained by mechanotransduction: cells constantly monitor the tension of their neighbours, and tension imbalance results in extrusion of abnormal cells (Wu et al., 2014). Notably, he discussed new results showing that caveolae in epithelial cells are necessary to maintain the normal level of tension that is required for the ability of these cells to extrude aberrant oncogene-transformed cells (Teo et al., 2019 preprint). Eduardo Moreno (Champalimaud Centre for the Unknown, Lisbon, Portugal) also discussed mechanical cell competition, focusing on molecular events in 'loser' clones. He showed that mechanically induced apoptosis relies on compression-driven inhibition of the EGFR/ERK pathway, leading to caspase activation (Moreno et al., 2019).

Combining quantitative 3D imaging and vertex models, Guillaume Salbreux (Francis Crick Institute, London, UK) showed how the morphology of epithelial pancreatic tumours can be predicted by the interplay of cytoskeletal changes in cancerous cells and local tubular geometry. This model is able to explain why exophytic lesions that expand outwards from the duct are only found in ducts below a critical diameter, whereas endophytic lesions are always found in larger ducts. Salbreux further confirmed and generalised these findings by documenting similar patterns of lesion growth in other tubular epithelial tissues, such as the liver and lung (Messal et al., 2019).

Although we may not always think of adult neurons as mechanically dynamic cell types, Miriam Goodman (Stanford University, USA) revealed that *C. elegans* sensory neurons undergo constant deformations – twisting, pushing and stretching – caused by body motions. Their axons and dendrites are protected from damage by their specialised and highly organised cytoskeleton, with a crucial role for specialised interaction partners of actin (spectrin) and microtubules (tau) (Krieg et al., 2017).

Mechanotransduction

The response of cells to mechanical forces often goes beyond passive deformation and frequently involves the activation of mechanosensitive biochemical pathways (Fernandez-Sanchez et al., 2015). Several talks reported on recent progress in understanding such mechanotransduction pathways, reinforcing known functions and uncovering novel functions for molecular mechanosensors, such as YAP/TAZ, lamins, transmembrane ion channels and Notch.

Mariaceleste Aragona (ULB, Belgium) discussed the ability of skin to expand in response to mechanical tension, a process that has long been known and exploited in reconstructive surgery, but the mechanisms of which remain unknown. Using lineage tracing in a mouse model of skin expansion based on self-inflating hydrogel patches inserted under the skin, she and her colleagues followed stem cell dynamics in stretched epidermis. Using an elegant

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mathematical model, they showed that tissue expansion is driven by an imbalance in cell fate decisions away from differentiation and toward self-renewal. Single cell RNA-seq and ATAC-seq further dissected the molecular mechanisms involved, showing that YAP/TAZ undergoes nuclear translocation upon stretching, downstream of Diap3 and actomyosin activity, and signals to the MAL/SRF pathway. Also focusing on YAP/TAZ, Hanna Engelke (LMU, Munich, Germany) presented recent progress toward gaining optogenetic control over the YAP/TAZ pathway. In her approach, she used a YAP transcription factor fused to a photoactivatable nuclear localisation sequence, which translocates to the cell nucleus under illumination, stimulating the expression of YAP target genes and cell proliferation, thus allowing growth to be controlled by light.

Dennis Discher's team (University of Pennsylvania, USA) investigated how nuclear integrity and DNA damage are affected by the mechanical properties of the extracellular matrix (ECM). They showed that acute perturbation of actomyosin contractility or ECM stiffness causes nuclear membrane rupture and DNA damage. This effect is exacerbated by deficiencies in lamin-A, which is normally stabilised and stiffens the nucleus in response to mechanical stress. The researchers concluded that lamin-A acts as a mechanosensitive 'mechano-protector' of genome integrity, particularly in highly active tissues such as heart or muscle (Cho et al., 2019).

The formation of cardiac valves in the heart is known to require mechanical signals from the incipient blood flow, but the exact mechanotransduction pathway remains unclear. Hajime Fukui (IGBMC, Strasbourg, France) used high-resolution live imaging of the zebrafish heart to show that endocardial cells transduce flow into calcium signalling, which, in turn, triggers valvulogenesis. Rashmi Priya (MPI, Bad Nauheim, Germany) also presented a study of zebrafish heart morphogenesis, using quantitative *in vivo* microscopy and genetic tools. She addressed how symmetry is broken in the initial myocardium monolayer to generate two distinct cell fates and a 3D patterned myocardial wall. She showed that heterogeneity in mechanical tension drives stochastic delamination of some cardiomyocytes from the outer compact layer to seed the inner trabecular layer. She further demonstrated that this heterogeneity arises in response to proliferation-induced tissue crowding, and that mechanics-induced delamination is sufficient to induce the Notch signalling pathway, which determines cell fate.

Finally, Prachi Richa (University of Göttingen, Germany) presented a surprising result: putative transmembrane channel-like (TMC) proteins, thought to be the mechanoreceptors of hair cells in the inner ear, are also involved in *Drosophila* embryonic morphogenesis, contributing to mechanotransduction between contractile amnioserosa cells during dorsal closure.

Force generation during morphogenesis

Control of morphogenesis by spatial patterns of gene expression

Thomas Lecuit (IBDM, Marseille, France), in his keynote lecture, summarised how patterned gene expression determines local force generation during morphogenesis, and, notably, how contractility in junctional and apical domains during early *Drosophila* development is controlled by distinct G protein-RhoGEF pathways (Garcia de las Bayonas et al., 2019). He then went to show that, reciprocally, mechanical forces trigger an active response in neighbouring cells during hindgut invagination: contractility is initiated transcriptionally in a small domain and then spreads posteriorly as a mechanically propagated wave (Bailles et al., 2019).

Enrico Coen (John Innes Centre, Norwich, UK) showed how spatial patterns of gene expression guide plant morphogenesis. Unlike animal cells, plant cells are incapable of directed migration, active contraction or neighbour exchanges, and morphogenesis thus proceeds strictly by differential growth. Using snapdragon flowers, Coen's team showed that orthogonal patterns of cell proliferation can generate out-of-plane deformation and the formation of a tissue-scale 'dome'. Orthogonal cell growth is oriented by polarised distribution of the auxin transporter PIN1 at the tissue scale, which is determined by dorsoventral gene expression. Coen then showed that other conflicting patterns of differential growth that create 'tissue conflicts' can generate a large variety of three-dimensional shapes and, as such, provide a flexible morphogenetic mechanism (Rebocho et al., 2017).

Finally, Daniela Panáková (MDC Berlin, Germany) showed how the planar cell polarity (PCP) pathway guides actomyosin contractility to convert the zebrafish heart from a tube into a two-chambered structure. She demonstrated that cardiac chambers are formed by topological rearrangements guided by PCP and upstream specialised Wnt ligands. The PCP pathway both guides cell neighbour exchanges, by patterning contractility, and triggers cardiac looping by restricting contractility to the apical side (Merks et al., 2018).

Emergent properties and self-organisation: from molecules to tissues

An emerging concept in the field is that of 'mechanochemical feedback' (Hannezo and Heisenberg, 2019) – the idea that the integration of biochemical and mechanical events at the tissue scale results in spontaneous emergent patterns that can only be understood by determining the combination and interaction of chemical and mechanical factors (Gilmour et al., 2017). Importantly, mechanochemical feedback can exist in the absence of any active mechanotransduction. Several talks illustrated this rising theme.

Two talks addressed one of the foundational problems of biological morphogenesis: the spatial distribution of diffusive factors that determine patterns – or morphogens – in tissues (Turing, 1952). First, Naama Barkai reviewed theoretical work on how a sharp peak of morphogen activity can be produced by a broad plateau of gene expression. The hypothesised mechanism involves 'morphogen shuttling', whereby other secreted proteins (such as inhibitors or proteases) interact with the morphogen to form complexes with variable levels of degradation, diffusion and biological activity. The resulting equilibrium determines the final distribution and activity of the morphogen itself (Eldar et al., 2002; Rahimi et al., 2019). Then, Adrien Hallou (University of Cambridge, UK) discussed a new mechanochemical model for self-organised pattern formation in multicellular tissues. Based on a biologically realistic description of multicellular tissues as active poroelastic media, the model overcomes the limitations of conventional reaction-diffusion models to show that mechanochemical coupling between morphogen concentrations, tissue mechanics and extracellular fluid flows provide alternative, Turing and non-Turing, mechanisms by which tissues can form robust spatial patterns of morphogens (Recho et al., 2019).

Five talks, all focusing on *Drosophila*, discussed how cell-cell interactions can yield cell or tissue shapes that cannot be explained by considering the individual cell level alone. First, Maria Leptin (EMBL Heidelberg, Germany) presented a detailed study dissecting how genetic and mechanical factors interact to define the shape of cells bordering the ventral furrow. Maithreyi Narasimha (TIFR, Mumbai, India) discussed work on epithelial tissue fusion (dorsal closure), showing that misalignments between cells and segments are corrected by controlled expansion-shrinkage of interfaces (Das Gupta and Narasimha, 2019). Yu-Chiun Wang (Riken CDB, Kobe,

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Japan) showed that robust formation of a linear cephalic furrow is guaranteed by mechanical coupling between neighbouring contracting cells, which buffers imprecisions and fluctuations in individual cell contractility (Eritano et al., 2019). Frank Jülicher (MPI Dresden, Germany) presented work on wing disc morphogenesis (Aigouy et al., 2010; Etournay et al., 2019). Using quantitative image analysis, he and his colleagues followed individual cell division, death, extrusion, shape changes and topological rearrangements during all pupal stages of development. They subsequently developed an elegant and minimal mechanical model for tissue dynamics that captures the essential physics of this morphogenetic process. Finally, Enrique Martin-Blanco (IBMB, Barcelona, Spain) discussed tissue replacement during metamorphosis, detailing the mechanical cell behaviours and molecular pathways that allow newly formed adult epithelial cells, generated by histoblasts, to invade and replace the larval epithelium (Ninov et al., 2007, 2010; Mangione and Martín-Blanco, 2018).

Finally, mechanochemical signals sometimes feedback onto morphogenetic processes themselves, to amplify or propagate them. Two elegant examples were provided, respectively concerning zebrafish gastrulation and ascidian neurulation. Carl-Philipp Heisenberg presented a live imaging study of the dynamics of tight junctions (TJs) between the enveloping cell layer and the yolk syncytial layer (YSL) in the gastrulating zebrafish embryo (Schwayer et al., 2019). The accumulation of Zonula Occludens-1 (ZO-1) in TJs scales with actomyosin network tension, demonstrating that TJs are mechanosensitive. This mechanosensitivity results both from a phase-separation mechanism and 'active gel' hydrodynamic instability. Specifically, non-junctional ZO-1 proteins form clusters through a liquid-liquid phase-separation mechanism. As these clusters bind to actin they are advected to TJs as a result of the higher actomyosin activity. Progressive accumulation of ZO-1 at TJs drives further retrograde actomyosin flow within the YSL in a typical positive-feedback loop mechanism. Edwin Munro (University of Chicago, USA) presented work that combined experiments with modelling to understand the unidirectional zippering of the neural tube in the chordate *Ciona intestinalis*. He showed that myosin II activity is triggered sequentially from posterior to anterior along the neural/epidermal (Ne/Epi) boundary by asymmetric localisation of cadherins and a RhoGAP, promoting local shortening of Ne/Epi junctions and thus driving the 'zipper' forward. Perhaps counter-intuitively, although directional progression of the contraction wave is mechanically driven, it does not require active mechanotransduction. Instead, local cell rearrangements behind the zipper allow transiently stretched cells to relax and permit zipper progression, while myosin II activation might be induced by signalling between cells brought into close proximity ahead of the zipper (Hashimoto et al., 2015; Hashimoto and Munro, 2019).

The cellular and molecular engines of morphogenesis: old and new players

Cells accomplish morphogenesis by generating forces in spatially controlled patterns. Classical examples of these 'morphogenetic engines' include patterned growth (often accompanied by cell division) and localised contractility; these 'old players' remained central to much of the research presented at the meeting. However, some unexpected factors also made recurrent appearances, reminding us that we should not naïvely assume our current repertoire of known morphogenetic mechanisms to be complete.

A fundamental mechanism that emerged from several of the studies presented was localised secretion or remodelling of the ECM. Maria-Carmen Diaz-de-la-Loza (Francis Crick Institute, London, UK) presented fresh insights into the function of one of the Hox genes, *Ultrabithorax* (*Ubx*). Although Hox genes have long been known to control segment morphology, we often still don't understand how. Her team's work showed that *Ubx* determines the difference between wings and halteres in *Drosophila* by triggering differential extracellular matrix remodelling through a secreted metalloproteinase (De las Heras et al., 2018). Pavel Tomancak (MPI Dresden, Germany) presented results on the newly discovered importance of locally expressed integrins, which mediate cell-ECM adhesion, during insect gastrulation (Münster et al., 2019). This research started from the observation that theoretical models of gastrulation in the beetle *Tribolium* were only able to account for known movements if one added one – then unobserved – factor: local adhesion to the vitelline membrane. Molecular studies confirmed that this hypothesised mechanism was real and implemented by a locally expressed, specialised integrin. This converged with Thomas Lecuit's lecture, supporting a pivotal role for localised integrin expression during *Drosophila* hindgut invagination.

Other studies revealed unexpected roles for what we thought were well-known molecules. An example is dynamin, famous for its function in endocytosis and more precisely in vesicle abscission. Elizabeth Chen (UT Southwestern Medical Centre, Dallas, USA), whose lab has contributed to demonstrating the role of actin in cell-cell fusion (Shilagardi et al., 2013), showed data supporting a role for dynamin in bundling actin inside the podosome-like structures that implement cell fusion (Zhang et al., 2019 preprint).

Finally, several talks highlighted how progress in imaging has catalysed the discovery of crucial functions for previously known, but somewhat neglected, cell structures. One example is provided by filopodia and microvilli – dynamic, finger-like, actin-filled protrusions that contribute to mesenchymal cell migration and axonal guidance, for example, but are rarely considered in the context of epithelial morphogenesis. For example, Timothy Saunders and his team (MBI, National University of Singapore) found an unexpected role for filopodia in the closure of the *Drosophila* heart tube, which involves specific adhesion and 'zippering' between cells of distinct subtypes. Here, cell-to-cell matching and closure appears to be mediated by specific adhesion between filopodia expressing complementary adhesion molecules (Zhang et al., 2018). In a similar discovery of unexpected active cell migration, Yohanns Bellaïche (Institut Curie, Paris, France) presented a study of the formation of the fold that delineates neck from thorax during *Drosophila* metamorphosis. Although neck invagination is an active process mediated by Myosin II contractility, laser ablation experiments show that it alone cannot explain cell flow in the thorax. Instead, thorax cells use cuticular ECM as a substrate for active migration.

On the origin of growth and form: the evolution of morphogenesis

Pavel Tomancak reminded the audience of the importance of an evolutionary framework for connecting, contextualising and comparing results obtained in diverse models in order to understand their history and deepen our understanding of biological phenomena. Following on from this, two talks tackled the ancient evolutionary ancestry of morphogenesis by comparing the mechanisms of animal and plant development to cellular processes in their single-celled or colonial relatives. First, Stephanie S.M.H. Höhn (University of Cambridge, UK) discussed an unusual process: a whole-embryo inside-out inversion that occurs during the development of the multicellular alga *Volvox*. With its round shape,

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Volvox resembles the abstract 'spherical cow' often postulated by physicists, and is thus exceptionally amenable to quantitative investigation. Höhn and co-authors leveraged this potential and, starting from spectacular light-sheet movies, established a mathematical model translating cell shape change into local curvature modification of an elastic shell (Haas et al., 2018). Second, Thibaut Brunet (University of California, Berkeley, USA) discussed the evolutionary origin of animal morphogenesis using a study of their closest living relatives, the choanoflagellates. He presented recent work on a newly discovered species, *Choanoeca flexa*, which forms cup-shaped multicellular sheets that use controlled, collective actomyosin contractility to invert their curvature quickly in response to light-to-dark transitions. This suggests that apical constriction evolved before multicellularity in the lineage that gave rise to animals, and supports a surprisingly deep evolutionary origin for this 'morphogenetic engine' (Brunet et al., 2019).

New techniques, models and systems

Much of the recent progress in our understanding of morphogenesis can be attributed to imaging that is highly resolved in space and/or time. Kate McDole (Janelia Farm, USA) presented a technical tour-de-force study in which light-sheet microscopy and adaptive optics allowed *in toto* live imaging of early development of mouse embryo, paving the way for deep learning to perform 3D image reconstruction, segmentation, cell tracking and fate mapping (McDole et al., 2018). Anne Herrmann (University of Cambridge, UK) presented work on the oscillatory movement of nuclei between the apical and basal surfaces of neuroepithelia, a process termed interkinetic nuclear migration (IKNM) (Azizi et al., 2019 preprint). Using a combination of light-sheet imaging of zebrafish embryonic retinas and mathematical modelling, she demonstrated that IKNM can be modelled as a diffusive process across a nuclear concentration gradient generated by the addition of new nuclei at the apical surface. She also discussed the potential relevance of this mechanism for understanding the stochastic cell fate decisions observed in neuroepithelia.

Beyond imaging, creative platforms to observe and manipulate morphogenesis *in vitro* were presented. Building up on his lab's earlier finding that antero-posterior elongation of the presomitic mesoderm (PSM) is driven by a gradient of random cell motility (Bénazéraf et al., 2010), Olivier Pourquié (Harvard Medical School, Boston, USA) and his team developed a microfabricated platform in which PSM explants are confined in PDMS (polydimethylsiloxane) channels of various sizes, allowing imaging of elongation and measurement of the stress generated by the elongating tissue. This showed that the PSM can elongate autonomously in an FGF-dependent fashion implemented by modulation of cell packing and extracellular space size. The relevance of these results was confirmed by cantilever-based measurements showing that the elongation stress generated *in vivo* (∼115 Pa) is similar to that *in vitro* (∼75 Pa).

Finally, organoids made an appearance in Qiutan Yang's talk (Liberali lab, FMI, Basel). She presented work combining organoid studies, single cell RNA-seq and vertex models to understand the molecular and cellular changes necessary for intestinal crypt formation in mice.

Conclusions

Given the breadth of discussions, new results, methods, theory and tools that were presented, one would certainly be convinced that this meeting was a success. In his seminal 1952 article 'The chemical basis of morphogenesis', Alan Turing wrote: 'The interdependence of the chemical and mechanical data adds enormously to the difficulty, and attention will therefore be confined, so far as is possible, to cases where these can be separated' (Turing, 1952). There was no doubt that the studies presented at this meeting have risen to Turing's challenge and overcome the dichotomy between biology and mechanics. By doing so, they have uncovered widespread and extensive cross-talk between mechanical and biochemical processes at all scales of biological organisation, thus justifying and reinforcing the need for integrative and interdisciplinary approaches.

Nevertheless, many challenges still lie in the way of fully understanding the mechanical determinants of morphogenesis. A true understanding of mechanobiology still awaits a precise and systematic understanding of the molecular building blocks that create, sense and effect mechanical forces inside cells, and how tissular and organismal properties such as biological forms emerge from cellular interactions. This will require designing new experimental approaches, combining *in toto* imaging at high resolution with spatio-temporally resolved molecular profiling of cells, as well as quantitative inference of tissue-level mechanical forces. In parallel, bottom-up approaches aiming at re-creating morphogenesis *in vitro*, such as organoid studies and synthetic biology experiments, will allow us to test our understanding of 'mechano-morphogenetic' processes in controlled settings. Analysing the data from these experiments will constitute a challenge both in terms of computational power and conceptual frameworks. In that respect, machine learning, information and graph theories are certainly appealing tools, and the day appears foreseeable when the power of these and other conceptual avenues could be harnessed to cut through complexity and eventually unravel the 'diagram of forces' that makes living beings.

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Competing interests

The authors declare no competing or financial interests.

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