

Mechanobiology of YAP and TAZ in physiology and disease

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Abstract | A growing body of evidence suggests that mechanical signals emanating from the cell's microenvironment are fundamental regulators of cell behaviour. Moreover, at the macroscopic scale, the influence of forces, such as the forces generated by blood flow, muscle contraction, gravity and overall tissue rigidity (for example, inside of a tumour lump), is central to our understanding of physiology and disease pathogenesis. Still, how mechanical cues are sensed and transduced at the molecular level to regulate gene expression has long remained enigmatic. The identification of the transcription factors YAP and TAZ as mechanotransducers started to fill this gap. YAP and TAZ read a broad range of mechanical cues, from shear stress to cell shape and extracellular matrix rigidity, and translate them into cell-specific transcriptional programmes. YAP and TAZ mechanotransduction is critical for driving stem cell behaviour and regeneration, and it sheds new light on the mechanisms by which aberrant cell mechanics is instrumental for the onset of multiple diseases, such as atherosclerosis, fibrosis, pulmonary hypertension, inflammation, muscular dystrophy and cancer.

Shear stress

The force vector component coplanar with a material cross section; a fluid moving along a solid boundary will generate a shear stress on that boundary.

TEAD factors

(TEA domain family members; also known as TEFs). A family of DNA-binding transcription factors that cannot induce target gene transcription on their own but need to associate with specific cofactors, such as YAP and TAZ.

A myriad of mechanical forces operate in a living body, including heart pumping, fluid shear stress, pressure and tensional forces in the skeletal system. In truth, physical forces affect every cell of our organs for the simple reason that our tissues have complex architectures, which are the product of an equilibrium of forces: internal pulling forces, dictated by the tension and organization of the cytoskeleton, and counterbalancing external forces, such as the topology and rigidity of the surrounding extracellular matrix (ECM) and other cells. As such, mechanical forces are informational systems by which cells perceive their position and shape and perturbations in their environment, inducing them to react by building, growing and healing tissues until a proper mechanical equilibrium is attained. Disturbance of these homeostatic mechanisms, caused by abnormal mechanical signals from the environment or cell-generated signals, is associated with a vast number of inborn or acquired diseases^{1–8}.

The profound influence of mechanical and physical constraints and forces on cell behaviour has long been recognized: for more than a century, before the advent of reductionist approaches in the late 1960s, cell and developmental biology were primarily explained in terms of the material properties and mechanical interactions between cells and tissues⁹. However, the lack of mechanistic, molecular understanding of these events sidelined this view of living systems, instead placing emphasis on signal transduction and genetics. The renaissance

of modern mechanobiology started with seminal discoveries on the mechanisms of mechanoperception at the level of cell–cell and cell–ECM adhesion sites^{7,10–13}, on the role of cytoskeletal proteins in these events, and by key demonstrations that cell mechanics and cell shape control cell proliferation, death and stem cell differentiation. That said, how mechanical forces, from the macroscopic to the microscopic scale, regulate cell fate by controlling gene expression remained a major black box in biology. This Review discusses how the identification of YAP (also known as YAP1) and TAZ (also known as WWTR1) as nuclear transducers of cell mechanics starts to fill this gap, linking the physicality of cells and tissues to potent transcriptional responses.

YAP and TAZ are transcriptional co-regulators that bind primarily to enhancer elements by using TEAD factors as DNA-binding platforms^{14–17}, an interaction originally described and functionally validated in *Drosophila melanogaster*^{18,19}. YAP and TAZ are at the nexus of multiple cellular inputs. They are best understood in the context of the Hippo signalling cascade, where the upstream kinases MST1 (also known as STK4) and MST2 (also known as STK3) trigger phosphorylation, and activation of the kinases LATS1 and LATS2, which in turn phosphorylate and inactivate YAP and TAZ by inducing their cytoplasmic retention and degradation. The reader can refer to excellent reviews for details on the Hippo and other pathways affecting YAP and TAZ activity^{20–23}. That said, a wealth of recent reports indicates that an

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Mechanotransduction

Mechanisms by which cells convert mechanical signals conveyed by the microenvironment into biochemical signals to adapt their behaviour to the environment.

overarching determinant of YAP and TAZ regulation is cellular mechanotransduction, operating primarily in a LATS-independent manner^{24–35}. Remarkably, YAP and TAZ read very diverse biomechanical signals and transduce them into biological effects in a manner that is specific for each type of cell and mechanical stress. As reviewed here, this includes cues emanating from ECM rigidity and topology, geometries of individual cells and whole tissues, stretching of epithelial monolayers in 2D and 3D, and tensional forces generated by asymmetric

cell division in early embryos, as well as diverse types of macroscopic forces, including stress generated by blood flow, muscle contraction and cyclic stretching of vascular smooth muscle cells (VSMCs). This ability to respond to the different mechanical inputs highlights the central role of YAP and TAZ as universal mechanotransducers and mechanoeffectors. Moreover, these roles of YAP and TAZ offer new means of interpreting and understanding classic aspects of tissue physiology and pathology in molecular terms. In this light, the discovery of YAP and TAZ as mechanotransducers is finally revealing the contribution of aberrant cell mechanics to the onset of multiple diseases, including atherosclerosis, fibrosis, cardiac hypertrophy, muscular dystrophy and cancer.

YAP and TAZ mechanotransduction

A major layer of regulation of YAP and TAZ occurs at the level of their subcellular distribution, as the activation of YAP and TAZ entails their accumulation in the nucleus. This cellular distribution is controlled by cell shape, by rigidity and topology of the ECM substrate^{24,27,36–39} and by shear stress^{40–42} (FIG. 1). For example, YAP and TAZ are localized in the cytoplasm in cells experiencing low levels of mechanical signalling, such as in rounded cells attached to a soft ECM (typically <1.5 kPa for epithelial cells) or to a small adhesive area (that is, 300 μm²); YAP and TAZ are instead nuclear in cells perceiving high levels of mechanical signalling, such as cells cultured on rigid substrates (typically at >5–10 kPa) or experiencing deformation and cytoskeletal tension (for example, cells stretching over an area of >3000 μm²), while remaining evenly distributed at more intermediate levels of mechanical signals^{24,27,43} (FIG. 1).

Moreover, YAP and TAZ are mechanically regulated by various regimens of cell stretching, such as deformations of epithelial monolayers, or by the rigidity and deformability of the ECM supporting cellular outgrowths in 3D, for example, in organoids^{24,27,44,45} (BOX 1; FIG. 1).

In all these contexts, YAP and TAZ are not just reading out cell and tissue mechanics but, crucially, also mediating the biological effects associated with mechanical signalling in each biological system, as detailed below (FIGS 2,3). Importantly, none of this could have been discovered without the development of new technologies and biomaterials that allowed first the study of mechanical signals independently from other variables *in vitro* and then the translation of this knowledge *in vivo*^{10,11,24,45,46}.

YAP and TAZ mechanotransduction requires integrity of the actin cytoskeleton, and YAP and TAZ activity is experimentally abolished by inhibitors of F-actin^{22,27,32,34,47} (FIG. 4). That said, regulation of YAP and TAZ by mechanotransduction is unrelated to the ratio of free G-actin to F-actin. For example, rounded cells contain more total F-actin than spread cells⁴⁸, and yet, in rounded cells, YAP and TAZ are inhibited. Rather, YAP and TAZ mechanotransduction relies on a specific organization of the F-actin cytoskeleton, as revealed by the potent inhibitory effects of F-actin capping proteins, such as capping actin protein of muscle Z-line (CAPZ), actin depolymerizing factor (ADF)/cofilin, on YAP and TAZ activity^{24,37,49},

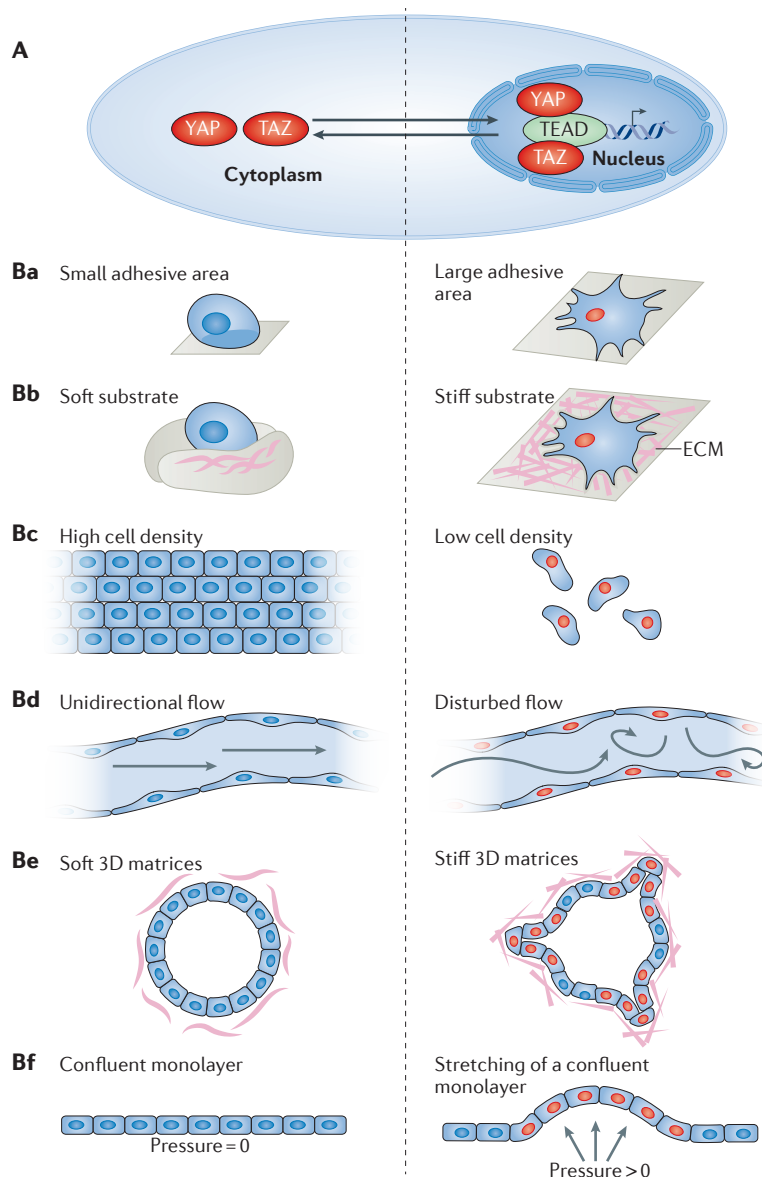


Figure 1 | Schematic representations of mechanical stimuli influencing YAP and TAZ subcellular localization and activity. **A** | When YAP and TAZ are mechanically activated (red), they translocate to the nucleus, where they interact with TEAD factors to regulate gene expression. **B** | Schematics illustrating how different matrix, geometry and physical conditions influence YAP and TAZ localization and activity: the left panels show conditions in which YAP and TAZ are inhibited and localized to the cytoplasm, whereas the right panels show conditions that promote YAP and TAZ nuclear localization (indicated by red colouring of cell nuclei). Part **Ba**, see REFS 24,27; part **Bb**, see REFS 24,27,36,43,72,76,105,106,108,109; part **Bc**, see REFS 24,75,91; part **Bd**, see REFS 40–42,83,84; part **Be**, see REFS 24,45,80; part **Bf**, see REFS 24,44. ECM, extracellular matrix.

Box 1 | YAP and TAZ mechanotransduction drives organoid biology

Organoids are 3D cultures of primary cells whose cellular composition and spatial organization closely recapitulate those of the tissue from which they derive (reviewed in REF 133). YAP and TAZ are essential for the maintenance and self-renewal of organoids derived from distinct tissues⁶⁵; regulation of organoids by cell mechanics is also instrumental for their ‘self-organizing’ and ‘self-renewing’ properties, that is, to preserve their stem cells and, at the same time, allow differentiation. Accordingly, YAP and TAZ are genetically required for the maintenance of intestinal stem cell function in organoids derived from adult intestinal crypts^{45,64,134}, and YAP was reported to be active in the growing (that is, ‘crypt-like’) compartment of the organoids, whereas it is confined to the cytoplasm of their more differentiated compartment⁴⁵. Typically, organoids are grown in Matrigel, a natural basement membrane substrate that is chemically and physically heterogeneous, making it impossible to dissect the relative contributions of distinct inputs on organoid biology. Intriguingly, when intestinal organoids are embedded in fully defined synthetic hydrogels, their growth is inhibited in soft substrates but dramatically fostered by more rigid matrices, a condition accompanied by, and requiring, nuclear accumulation of YAP and TAZ⁴⁵ (FIG. 1). As cells accumulate and the organoid expands, increased cell crowding progressively limits YAP and TAZ activity (FIG. 1), promoting differentiation. As such, to support long-term organoid expansion, colonies must be transferred into a cleavable, and thus mechanically dynamic, artificial matrix that allows budding of ‘crypt-like’ finger-shaped protrusions at whose tips stem cells are preserved by nuclear (active) YAP and TAZ⁴⁵.

As another example, in human airway organoids, YAP and TAZ are nuclear in basal (progenitor) cells and absent in differentiated cells. Similarly to intestinal organoids, mechanoactivation of YAP and TAZ by raising tension in the F-actin cytoskeleton is essential for branching morphogenesis of these lung organoids *in vitro*¹³⁵.

Capping proteins

Proteins (including CapZ but also ADF/cofilin, which are also F-actin-severing proteins) that bind to the growing (barbed) end of F-actin filaments, blocking the binding of other actin subunits to the growing filament.

Vinculin

A focal adhesion protein that links the cytosolic domain of integrins to the F-actin cytoskeleton.

Talins

Focal adhesion proteins that link integrin to the F-actin cytoskeleton either directly or indirectly (through binding to vinculin or α -actinin).

Focal adhesion kinase

(FAK). A cytosolic tyrosine kinase associated with focal adhesions and required for integrin-dependent cell motility, cell spreading and cell survival.

SRC-family kinases

A family of tyrosine kinases (composed of nine different members: SRC, YES, FYN, FGR, LCK, HCK, BLK, LYN and FRK) that can target cytosolic, nuclear or membrane proteins, having a broad impact on the phosphotyrosine proteome and on diverse cell behaviours.

and similar regulatory effects of other F-actin-regulatory proteins, such as angiomin^{50,51}, Spectrins^{52,53} and the tight junction protein zonula occludens protein 2 (ZO2)^{54–58} (FIG. 4). In addition, actin contractility and increased tension of actin cytoskeleton have been linked to YAP and TAZ activation^{24,27}. However, the exact mechanisms of how the actin cytoskeleton impacts YAP and TAZ signalling in the context of mechanotransduction are not known.

Once mechanical forces are detected at the cell surface, primarily at integrin–ECM and cell–cell adhesion sites, the information they carry must be propagated and preserved within the cytoskeleton before force dissipation^{59–61}. This is mediated by mechanosensory systems that include integrins and adherens junctions, adaptor proteins such as vinculin and talins, and focal adhesion kinase (FAK; also known as PTK2) and the SRC-family kinases that connect the extracellular mechanical world to the F-actin cytoskeleton^{62,63}. Notably, these mechanosensory proteins have been implicated in YAP and TAZ regulation^{28,33} (FIG. 4).

Finally, cells are not passive force receivers but dynamically respond to external forces with a proportional cell-generated force. This occurs through assembly of an actomyosin contractile network and cytoskeletal remodelling¹. Consistently, in some cell types, YAP activation by a stiff ECM can be blunted by inhibitors of myosin light chain kinase (MLCK), non-muscle myosin II or RHO-associated protein kinase (ROCK)²⁷. Moreover, integrin activation restructures the cytoskeleton by activating RHO GTPases. Consistently, RHO signalling is essential for YAP and TAZ activity, and this has been experimentally exploited in various systems, either genetically or by using RHO inhibitors^{24,27,32,34,47} (FIG. 4; see also below).

Stem cell mechanobiology

In adult tissues, nuclear YAP and TAZ are typically found in locations enriched for somatic stem cells or progenitors, such as at the bottom of intestinal crypts, in cells of the basal layer of the skin, or in basal and luminal mammary gland progenitors^{57,64,65}. YAP and TAZ are remarkably essential for tissue repair *in vivo*⁶⁶, as well as for the growth of organ-specific stem cells *ex vivo* as organoids (BOX 1). By contrast, they appear dispensable for normal homeostasis of most adult epithelial organs⁶⁶. These seemingly contradictory observations may be reconciled if one considers that, under homeostatic conditions, adult organs may keep their cell mechanics below the threshold required to activate the transcriptional effects of YAP and TAZ. Recent results on mutant mice lacking ADF/cofilin in the skin and liver, where extensive cytoskeletal remodelling and F-actin accumulation are accompanied by induction of massive organ overgrowth occurring in just a few days (phenocopying the effects of YAP and TAZ activation)⁶⁷, are consistent with the view that the levels of mechanical tension and cytoskeletal organization of normal epithelial tissues are insufficient to sustain YAP and TAZ responses.

YAP and TAZ mechanobiology in embryonic stem cells.

YAP and TAZ and their regulation by cell mechanics control the first cell fate decision in the mammalian embryo, namely, the specification into cells of the trophoblast or cells of the inner cell mass (ICM). YAP and TAZ are required for the adoption of the trophoblast fate, and experimental overactivation of YAP and TAZ in ICM cells is sufficient for them to acquire trophoblast markers⁶⁸. Consistent with this, in a normal blastocyst, YAP and TAZ are nuclear in trophoblast cells and cytoplasmic in ICM cells. The Hippo kinases LATS1 and LATS2 are relevant to blunting YAP and TAZ activity at these stages; yet, it remains unclear whether Hippo signalling itself is primarily involved in patterning YAP and TAZ in the blastocyst or whether it provides an inhibitory signal setting a threshold above which the trophoblast fate is induced by other patterning signalling. Strikingly, recent work highlighted cell contractility and cell shape as signals that regulate YAP and TAZ in the early mouse embryo. Contraction of ICM cells forces the outer cells to stretch over the external surface of the embryo, fixing their fate as trophoblast cells. Inhibition of such pulling forces by exposing mouse embryos to blebbistatin (an inhibitor of non-muscle myosin II and actomyosin contractility) or to ROCK inhibitors abolishes nuclear localization of YAP in outer cells and their trophoblast commitment^{69,70} (FIG. 5a).

YAP and TAZ are required to foster survival and prevent differentiation of human embryonic stem cells (hESCs), allowing their long-term propagation^{71–73}. YAP and TAZ are localized to the nucleus in hESCs cultured under standard conditions on stiff substrates, but they relocalize to the cytoplasm when cells are forced to adopt a rounded shape by low levels of mechanical signals. Consistent with YAP and TAZ inhibition, hESCs differentiate in these mechanically inhibited

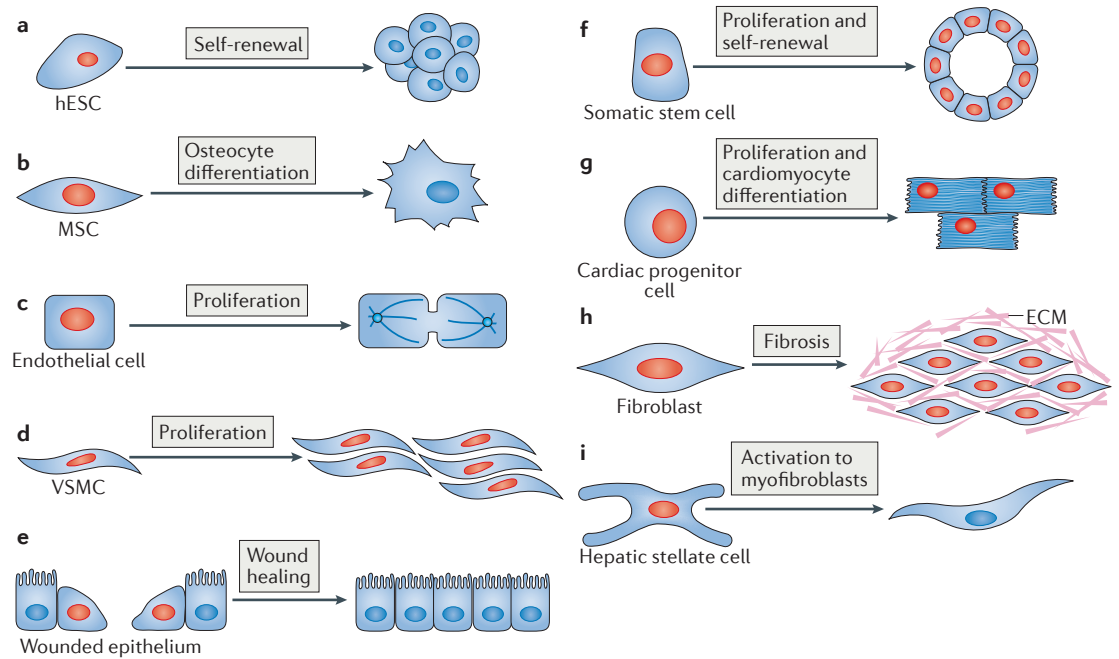


Figure 2 | Biological responses caused by nuclear accumulation and activation of YAP and TAZ by high levels of mechanical signalling. When YAP and TAZ are activated by increased mechanical inputs (see FIG. 1, right panels), this causes various YAP-dependent and TAZ-dependent biological effects (indicated in boxes) that are specific for each cell type. Red-labelled nuclei indicate the nuclear activation of YAP and TAZ. Part **a**, see REFS 71–73; part **b**, see REFS 27,80,82–84; part **c**, see REFS 27,40–42,105,106; part **d**, see REFS 102,103,105; part **e**, see REFS 43,93,94,99,100,115; part **f**, see REFS 28,45,64,65; part **g**, see REF. 98; part **h**, see REFS 105,110–112; part **i**, see REFS 108,109. ECM, extracellular matrix; hESC, human embryonic stem cell; MSC, mesenchymal stem cell; VSMC, vascular smooth muscle cell.

RHO-associated protein kinase (ROCK). A serine/threonine kinase that is best understood as a downstream effector of the small GTPase RHOA. ROCK favours formation of stress fibres and contractility of the actomyosin cytoskeleton by phosphorylating LIM domain kinase and myosin light chain kinase.

Intestinal crypts
Simple tubular invaginations at the base of intestinal villi (also known as crypts of Lieberkühn). Intestinal crypts are lined by various cell types: goblet, enteroendocrine, tuft, Paneth cells and intestinal stem cells.

Trophoblast
The outer layer of a blastocyst, composed of polarized cells from which part of the placenta originates.

Inner cell mass (ICM). A mass of cells lying on one side of the blastocyst cavity from which all the adult tissues eventually derive.

Blastocyst
A structure formed during the development of mammalian embryos after the morula stage and before implantation, and is composed of an external layer (trophoblast) surrounding an internal cavity (blastocoel) and the inner cell mass.

Elastic modulus
A measure of the resistance of an object to elastic deformation.

conditions^{71,72,74–76} (FIGS 2,3). On the basis of these reports, it is tempting to speculate that differentiation of pluripotent ESCs might be directed towards specific cell fates by gradients of mechanical signals quantitatively tuning the strength and duration of YAP and TAZ activity, in a manner conceptually similar to the effects of gradients of soluble growth factors or morphogens that are thought to pattern the early embryo.

YAP and TAZ mechanotransduction guides mesenchymal stem cell differentiation. Mesenchymal stem cells (MSCs) are a paradigm model system in the mechanotransduction field, and it has been shown that differentiation of MSCs along distinct cell fates is actually dictated by the physical features of the cell micro-environment rather than by soluble signals⁷⁷. This can be experimentally controlled by allowing cells to stretch on stiff substrates or, conversely, by inducing cell rounding either by confining cells to small adhesive areas or by culturing them on soft substrates (FIG. 1).

Human MSCs (hMSCs) adopt an osteogenic fate when allowed to stretch (FIG. 2), whereas they turn into adipocytes at low mechanical stresses (FIG. 3). Treatments with inhibitors of actomyosin contractile units block osteogenesis and favour adipogenesis, indicating that mechanically driven cell fate determination relies on the contractility of the actomyosin cytoskeleton. Conversely, overexpression of constitutively active ROCK forces osteogenic differentiation in cells plated on small fibronectin islands⁷⁷. In another study, it has

been demonstrated that hMSC differentiation could be regulated by the elasticity of the underlying substrate¹¹. By matching the elastic modulus of distinct tissues, it was shown that MSCs adopt neurogenic, myogenic or osteogenic fates at increasing substrate rigidities (FIG. 5b). This occurs independently from soluble cues, but is dependent on the degree of hMSC spreading and cytoskeletal contractility.

Despite these elegant bioassays, the nuclear effectors of MSC mechanosensitivity remained elusive until the identification of YAP and TAZ²⁷. Previous work had shown that TAZ was required for osteogenic differentiation of MSCs⁷⁸. Now, it is known that YAP and TAZ transcriptional activity couples substrate compliance to hMSC differentiation, as YAP and TAZ depletion impairs osteogenic differentiation on stiff substrates, skewing MSCs towards adipogenic differentiation; conversely, overexpression of a hyperactive form of YAP is sufficient to redirect hMSCs towards an osteogenic fate on soft substrates²⁷. Importantly, YAP and TAZ are genetically required for proper osteogenic differentiation, as *Yap*^{+/-}/*Taz*^{+/-} MSCs display impaired osteogenesis despite being cultured under permissive conditions⁷⁹.

YAP and TAZ also act as key mechanotransducers in specialized MSCs, such as in skeletal progenitors (or skeletal stem cells (SSCs)), when these cells are embedded within 3D collagen gels⁸⁰. ECM remodelling is well known to occur during morphogenesis and wound healing; the discovery of YAP and TAZ allows linking

Enamel

One of the four different tissues that compose the tooth; it is the outermost, highly mineralized tissue formed by epithelial cells known as ameloblasts.

such remodelling to nuclear responses. Membrane type 1-matrix metalloproteinase (MT1-MMP; also known as MMP14) is key to remodelling collagen fibres in the 3D environment of SSCs, in turn promoting cytoskeletal tension through a RHOA–ROCK pathway and acquisition of a spread cell shape, which are essential for YAP and TAZ activation. Indeed, *Mmp14*^{-/-} mouse SSCs display impaired osteogenic differentiation and enhanced adipogenic differentiation, both *in vitro* and *in vivo*, in a manner that is genetically dependent on YAP and TAZ dosage⁸⁰ (FIG. 5c).

Mechanical regulation of hMSC fate is not only dependent on the physical properties of the underlying substrates but also modulated by physical contacts with other cells. During development, MSCs engage in cell–cell and cell–ECM contacts simultaneously, raising questions about how cells integrate distinct mechanical inputs. This has been addressed in elegant *in vitro* experiments that indicated that the engagement of N-cadherin (also known as cadherin 2; involved in cell–cell interactions) reduced the contractile state and thereby the nuclear YAP and TAZ localization in MSCs, resulting in altered interpretation of ECM rigidity⁸¹.

MSCs can be mechanically regulated also by the shear stress generated in tissues by extravascular liquid flow. Culturing MSCs on microfluidic chips that mimic interstitial shear stress increases YAP and TAZ activity in a RHO–ROCK-dependent manner, enhancing osteogenic and fibrochondrogenic differentiation^{82–84}. Moreover, TAZ-dependent mechanotransduction has been proposed as the mechanism responsible for the microgravity-induced bone loss observed in space flights; culturing MSCs under microgravity indeed resulted in blunted TAZ nuclear accumulation and reduced osteogenesis⁸⁵.

Integrin–FAK signalling to YAP and TAZ in tooth progenitors. The mouse incisors grow continuously, in part through constant deposition of enamel by epithelial cells generated from a pool of progenitor cells. This pool needs to expand before differentiation, and high nuclear YAP and TAZ activity is instrumental for this process. YAP and TAZ activation in this context requires signalling from the surrounding ECM transduced by $\alpha 3$ integrin and FAK and is LATS independent²⁸.

YAP, muscle stem cells and muscle hypertrophy. Muscles grow in response to physical exercise and degenerate when muscle contraction is impaired. This is regulated in a YAP-dependent manner. In mouse models, mechanical overload sustains YAP activity. This overload, in turn, can induce hypertrophy and is required to protect against atrophy caused by denervation. In skeletal muscle cells cultured *in vitro*, high YAP activity promotes proliferation of muscle stem cells, whereas YAP inactivation by soft substrates fosters myogenic differentiation⁸⁶. The underlying mechanisms are only partially understood: in the chick embryo, contraction of postmitotic muscle fibres led to nuclear YAP, in turn leading to activation of Notch signalling in adjacent muscle progenitor cells, which prevented their differentiation⁸⁷ (FIG. 5d). Moreover, muscle size is known to be controlled by transforming growth factor- β (TGF β)–myostatin–SMAD signalling and by the mTOR–AKT cascade, which, in other contexts, have been shown to crosstalk with YAP and TAZ activity^{28,88–90}. This raises the intriguing possibility that, in the skeletal muscle, YAP and TAZ may relay mechanical signals to other transcription factors or metabolic pathways, as such harmonizing complex cell behaviours to the mechanical state of the cell.

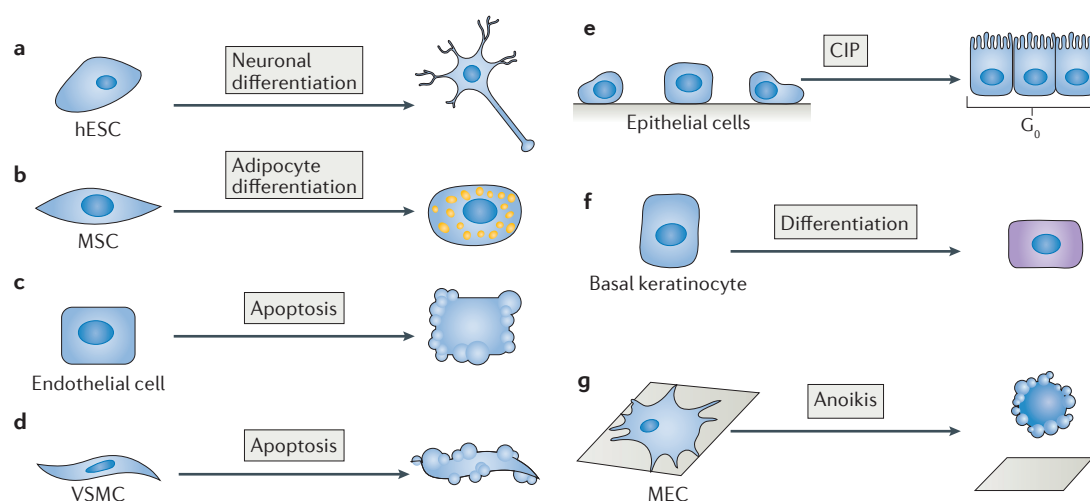


Figure 3 | Biological responses caused by cytoplasmic YAP and TAZ retention and inhibition due to low levels of mechanical stimuli. Low levels of mechanical forces (see FIG. 1, left panels) blunt YAP and TAZ activity, and this results in various YAP-dependent and TAZ-dependent biological effects in diverse cell types. Part **a**, see REFS 71,72,74–76; part **b**, see REFS 27,80,81,85; part **c**, see REF. 27; part **d**, see REFS 102,103; part **e**, see REFS 24,91; part **f**, see REFS 43,93,94,99,100,115; part **g**, see REF. 47. CIP, contact inhibition of proliferation; hESC, human embryonic stem cell; MEC, mammary epithelial cell; MSC, mesenchymal stem cell; VSMC, vascular smooth muscle cell.

Contact inhibition of proliferation

A classic ‘social-like’ behaviour of epithelial cells is contact inhibition of proliferation (CIP), whereby the tissue stops growing after reaching a critical cell density. CIP is transiently relieved at the margin of a wound after tissue damage and is more permanently overridden in tumours⁶⁶. CIP can be molecularly explained by inhibition of YAP and TAZ⁹¹, simply because crowding confines cells to small areas, putting them into a low contractility and YAP and TAZ repressive cytoskeletal states²⁴ (FIG. 3). To a substantial extent, this occurs in

a LATS-independent manner²⁴. Consistent with CIP being a mechanically regulated biological response, CIP is overcome by ECM stiffening⁹², by loss of actin capping and severing proteins, or by cell stretching^{24,44} processes that activate YAP and TAZ transcription (FIG. 1). More generally, CIP provides a proof of principle that cells can read patterns of mechanical forces associated to specific 2D topologies and 3D architectures of living tissues, turning them into a pattern of YAP and TAZ activity that can then guide specific cell behaviours.

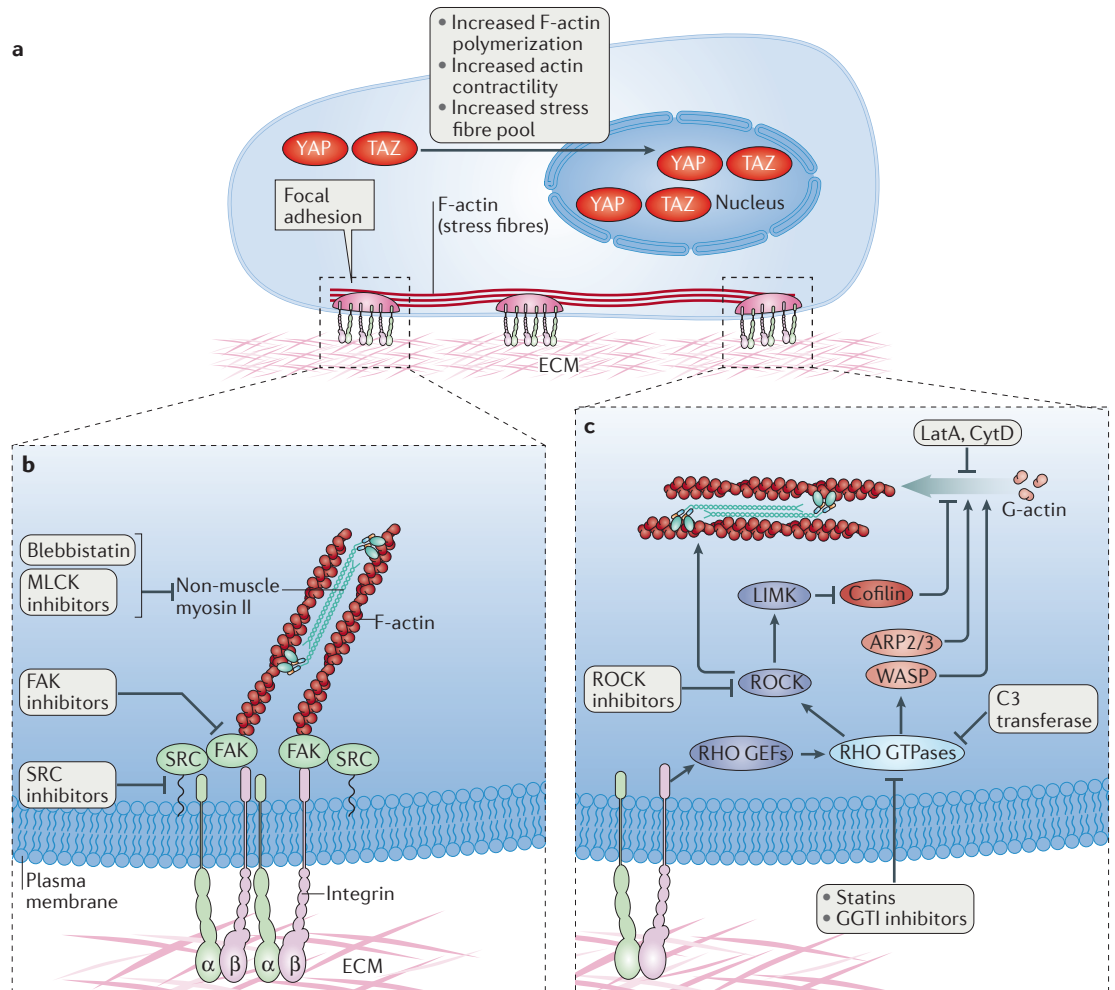


Figure 4 | Molecular players involved in YAP and TAZ mechanotransduction. a | Cell–extracellular matrix (ECM) adhesion complexes undergo force-dependent conformational changes to trigger an increase in actin polymerization and a consequent increase in stress fibre contractility, resulting in YAP and TAZ activation. **b** | A stiffer matrix causes integrin clustering, which results in the activation of focal-adhesion-associated kinases, such as focal adhesion kinase (FAK) and SRC, which in turn favour stress fibre growth, stability and contractility, thereby activating YAP and TAZ^{28,33,59,62,63,114}. SRC has also been shown to phosphorylate YAP, and this was linked to YAP activation downstream of SRC. However, whether such phosphorylation events by SRC are indeed causal for effective YAP (and TAZ) activation remains to be determined. **c** | Signalling from focal adhesions activates RHO GTPases, which can directly activate actin-regulatory proteins, such as actin-related protein 2/3 complex (ARP2/3) and Wiskott–Aldrich syndrome protein (WASP), or can favour F-actin polymerization through the activation of RHO-associated protein kinase (ROCK); active ROCK promotes actomyosin contractility and activates LIM domain kinase (LIMK), which in turn inhibits the F-actin-severing protein cofilin. In part **b** and part **c**, relevant chemical inhibitors of key players in the YAP and TAZ mechanotransduction pathway are shown (grey boxes)^{24,27,32,34,47}. C3 transferase, *Clostridium botulinum* C3 exoenzyme; CytD, cytochalasin D; GEFs, guanine nucleotide exchange factors; GGTI, geranylgeranyl transferase I; LatA, latrunculin A; MLCK, myosin light chain kinase.

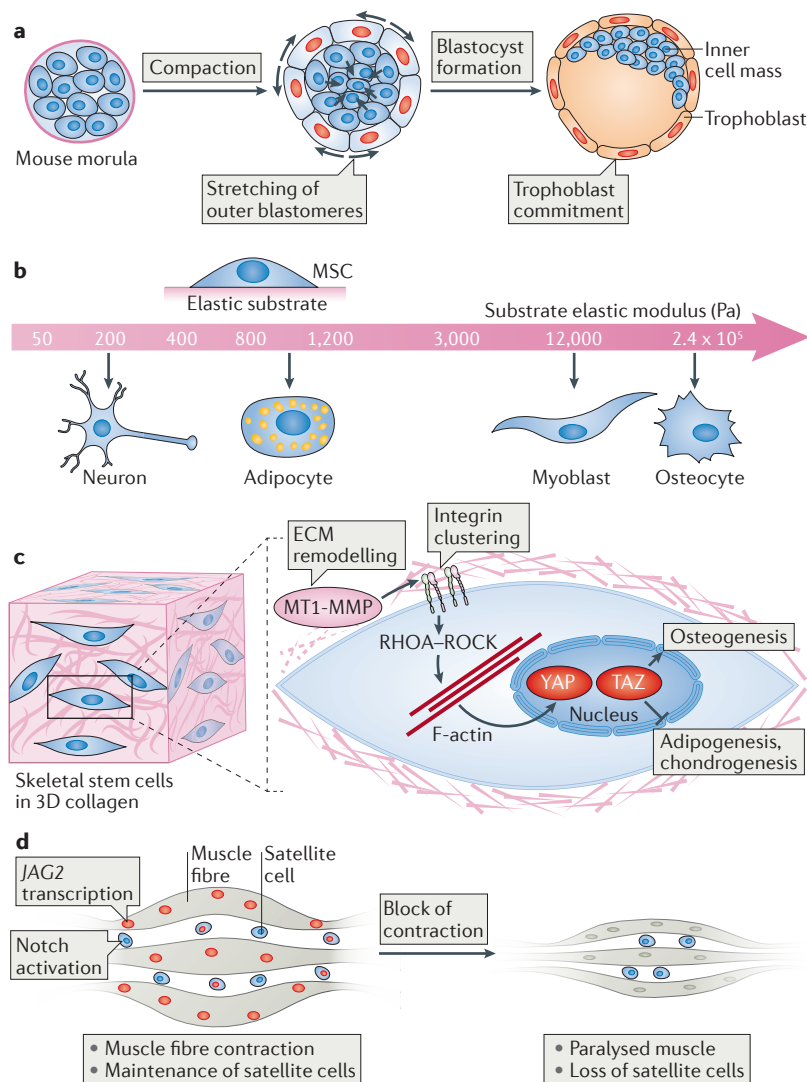


Figure 5 | YAP and TAZ mechanotransduction in stem cell biology. **a** | Trophoblast commitment in the early mouse embryo involves a phase of stretching of the outer cells caused by compaction (inward-pointing arrows) of the embryo. This stretching causes nuclear translocation of YAP and TAZ in the outer cells and the acquisition of a trophoblast fate. Cells that do not activate YAP and TAZ become the cells of the embryo proper (the inner cell mass)^{69,70}. **b** | *In vivo*, cells experience a range of different extracellular matrix (ECM) elasticities in different tissues. By recapitulating these different stiffnesses *in vitro*, it was found that mesenchymal stem cells (MSCs) differentiate optimally into neurons, adipocytes, skeletal muscle cells or osteoblasts at specific elasticities that match the physiological ECM stiffness of their corresponding natural niche. **c** | Control of skeletal stem cell lineage commitment mediated by ECM remodelling by membrane type 1-matrix metalloproteinase (MT1-MMP). By affecting the ECM and thereby the cell shape of MSCs in a collagen-based 3D environment, MT1-MMP promotes integrin clustering and the concomitant activation of the integrin–RHOA pathway, thereby triggering YAP and TAZ activation, which switches on a programme that favours osteogenic differentiation over alternative adipogenic and chondrogenic cell fates⁸⁰. **d** | YAP-dependent and TAZ-dependent maintenance of satellite cells (muscle progenitors) by muscle contraction. In the chick embryo, contraction of postmitotic muscle fibres triggers YAP-dependent expression of the Delta-like ligand JAG2, which activates Notch in muscle progenitor cells and prevents their differentiation. Blocking embryonic muscle contraction (muscle paralysis) inhibits YAP and transcription of JAG2 in myofibres, causing a shift towards differentiation of satellite cells. Enhanced differentiation eventually depletes the pool of muscle progenitor cells and interferes with regenerative potential of the tissue⁸⁷. Similarly, expression of Delta-like ligands, such as DLL1 and JAG2, is also downstream of YAP and TAZ in epidermal stem cells⁴³.

Mechanoregulation of regeneration

The healing of damaged tissues entails several steps, including changes in the stiffness of surrounding ECM and increased cellular mechanoresponsiveness in the cells bordering the wound. This may lead to dedifferentiation, transdifferentiation and acquisition of migratory and/or progenitor-like properties. A causal relationship between changes in the mechanical properties of a tissue and YAP and TAZ activation during regeneration has been proposed in several tissues (FIG. 2). One example is YAP-driven lung regeneration caused by surgical removal of a part of lung tissue (pneumonectomy)⁹³. Pneumonectomy induces expansion of the remaining lung epithelia by activating latent stem cell properties of alveolar type II (AT2) cells, which eventually proliferate and differentiate into AT1 cells, the main cell type of the alveolar epithelium. Pneumonectomy induces stretching and actin polymerization in AT2 cells through a mechanism that depends on the RHO GTPase CDC42. In turn, cell stretching promotes YAP nuclear localization and transcriptional activities (FIG. 2), which are required for the expansion of AT2 cells and replenishment of AT1 cells. Accordingly, genetic deletion of CDC42 or YAP impairs regeneration after pneumonectomy⁹³.

Skin injury represents another model for YAP-dependent and TAZ-dependent regeneration. The epidermis is a classic mechanosensitive tissue: upon stretching — such as during growth-associated post-natal development or pregnancy — it adapts to body expansion by increasing its own size through an elevated proliferation of the basal cell layer, containing epidermal progenitors. Wound repair occurs through activation of basal keratinocytes at the margin of the wound; these cells migrate towards the injured area and proliferate to mediate wound re-epithelialization. YAP and TAZ are activated in basal keratinocytes near the wound (FIG. 2) and are required for proper wound closure⁹⁴. How YAP and TAZ are activated in the proximity of the wound margins remains only partially understood, but at least *in vitro*, ‘wounding’ a cell monolayer results in a localized loss of cell polarity, loss of contact inhibition (see above) and cell stretching (FIG. 2), whereas *in vivo*, increased rigidity of the underlying dermis occurs — all conditions associated with mechanical activation of YAP and TAZ. Intriguingly, loss of $\beta 1$ integrin results in impaired wound closure similar to YAP and TAZ knockout⁹⁵.

Indeed, mechanical signals, such as cell spreading and ECM rigidity, can activate YAP and TAZ in basal keratinocytes (FIG. 2), and this is required for preserving epidermal stemness (FIG. 3), which occurs through the suppression of the Notch pathway, a potent inducer of epidermal differentiation and stratification⁴³. Consistently, unscheduled activation of actomyosin-mediated cellular tension in the skin of transgenic mice by expressing ROCK2 induces epidermal hyperplasia in a manner that is attenuated by inhibition of FAK, LIM domain kinase (LIMK1) or myosin ATPase⁹⁶.

Mechanically regulated YAP and TAZ activity may also have a role in cardiac regeneration. Myocardial infarction is accompanied by a prominent remodelling and stiffening of the myocardial ECM⁹⁷, and YAP

and TAZ are expressed and localized in the nucleus of murine cardiac cells at the infarction border zone⁹⁸. YAP and TAZ have key functions in adult cardiac progenitor mechanosensory responses, being instrumental for cardiomyocyte proliferation at specific rigidities⁹⁸. YAP activation indeed increases cardiomyocyte proliferation, reduces scar size and improves cardiac regeneration in a myocardial infarction model^{99,100} (FIG. 2). Thus, activation of YAP through biomaterial design may offer new means of promoting cardiac repair after injury.

Mechanotransduction in diseases

Aberrant mechanical signalling has been associated with the pathogenesis of multiple diseases, and this can be caused by changes in the physical and structural features of the cell microenvironment or by defects in how cells perceive mechanical inputs¹⁻⁸. Strikingly, most of these defects eventually affect cell behaviour through YAP and TAZ deregulation.

Atherosclerosis and cardiovascular diseases. YAP and TAZ are key factors in the pathophysiology of the cardiovascular system, whose development and homeostasis are intimately connected to mechanical forces associated with blood flow, and for which defects in mechanical parameters are often the primary initiators of disease³. Endothelial cells are a convenient model in mechanobiology. These cells experience vascular shear stress, and blood pressure and their behaviour is under the control of cell mechanics; by using small and large microprinted adhesive areas, it has been shown that individual endothelial cells can be induced to proliferate or die depending on cell shape, that is, by the degree of cytoskeletal distortion and reorganization allowed by the geometry of their environment¹⁰.

YAP and TAZ knockdown impairs proliferation of endothelial cells that are allowed to stretch over big microprinted islands, whereas YAP overexpression inhibits apoptosis and rescues proliferation of endothelial cells seeded on small islands²⁷ (FIGS 2,3). This prompted further studies showing that YAP and TAZ mechanotransduction mediates the pathological effects of disturbed blood flow on atherogenesis³. YAP and TAZ are inhibited in endothelial cells cultured under static fluid flow (which is atheroprotective), whereas they localize to the nucleus when cultured under disturbed (oscillatory) flow conditions (which mimic atherogenic mechanical stress)^{40,41} (FIG. 1). Under these *in vitro* conditions, nuclear YAP and TAZ foster endothelial cell proliferation and expression of adhesive factors for monocytes, correlating with key pathogenic events in atherosclerosis, such as endothelial thickening and recruitment of monocytes, which eventually turn into plaque-forming foam cells.

YAP is also genetically required for this atherogenic response of endothelial cells *in vivo*⁴¹, thereby offering an unexpected therapeutic opportunity for managing atherosclerosis. The main anti-atherosclerotic drugs are statins, which are cholesterol-lowering compounds and are commonly used as first-line therapy for patients with cardiovascular diseases; however, the cholesterol

biosynthetic pathway that is targeted by statins also generates the geranylgeranyl lipid intermediates that anchor RHO GTPases to the plasma membrane and are essential for cytoskeletal tension and remodelling in the YAP and TAZ mechanotransduction pathway³² (FIG. 4). Consequently, the anti-inflammatory and anti-plaque effects of statins are now being re-interpreted as being mediated, at least in part, by their capacity to inhibit YAP and TAZ⁴⁰. More broadly, this suggests that YAP and TAZ mechanotransduction could be specifically targeted for improved atherosclerosis treatment.

Recent work on Yap1-null zebrafish embryos showed vessel regression in mutant larvae, with vessel stenosis and retraction between 4.5 and 6.5 days post-fertilization. This phenotype can be associated with the requirement of YAP and TAZ activation by shear stress and actomyosin contractility in endothelial cells for vasculogenesis at the onset of blood circulation⁴².

Apart from endothelial cells, mechanically regulated YAP and TAZ activity plays a part in VSMCs for vessel development and homeostasis. *Yap* knockout in smooth muscle cells in mice results in embryonic lethality, possibly owing to abnormal aortic development¹⁰¹. YAP and TAZ also drive proliferation and migration of VSMCs (FIG. 2) in the context of vessel restenosis after coronary angioplasty. These responses can be blunted by inhibition of RHO, ROCK or F-actin remodelling, which interfere with YAP and TAZ activation and result in VSMC apoptosis^{102,103} (FIGS 3,4).

Pulmonary hypertension is a deadly vascular disease, characterized by increases in proliferation of endothelial cells and VSMCs and by abundant deposition and stiffening of the vascular ECM¹⁰⁴. It has been recently proposed that YAP and TAZ activation downstream of ECM stiffening serves as a molecular driver of pulmonary hypertension^{105,106}. Indeed, a stiff ECM activates YAP and TAZ *in vivo* and *in vitro*, triggering proliferation of adventitial fibroblasts, endothelial cells and VSMCs; in turn, YAP and TAZ increase ECM deposition and remodelling via collagen synthesis, lysyl oxidase (LOX) production and secretion of pro-fibrotic factors¹⁰⁵, all in all fuelling a YAP and TAZ mechano-active feedback system. Of note, YAP and TAZ mechanotransduction may contribute to well-established metabolic dysfunctions associated with pulmonary hypertension, including increased glycolysis at the expense of mitochondrial oxidative phosphorylation because YAP and TAZ directly control the transcription of key metabolic enzymes that are responsible for glycolysis and glutaminolysis^{105,106}. Importantly, reduction of ECM stiffening with LOX inhibitors blunted nuclear YAP and TAZ and ameliorated the end-stage manifestations of pulmonary hypertension in murine models¹⁰⁶. It is tempting to propose that targeting mechano-activated YAP and TAZ and/or their downstream effectors in pulmonary hypertension could be a valuable tool to treat patients affected by this devastating disease.

Tissue fibrosis. Fibrotic diseases, which include liver cirrhosis, pulmonary, kidney and cardiac fibrosis, and systemic sclerosis, account for at least one-third of

LIM domain kinase

An actin-bound kinase that phosphorylates and inactivates actin-binding and actin-severing proteins, such as ADF/cofilin, thereby fostering the stabilization of F-actin filaments. LIM domain kinases are activated by RHO GTPases.

Foam cells

Lipid-filled M2 macrophages that form when monocytes are recruited to an atherogenic area of the arterial wall and contribute to the formation of the atherosclerotic plaque; these cells are derived from monocytes that engulf lipoprotein particles.

Vessel stenosis

Abnormal narrowing of a blood vessel (might be referred to as coarctation in the case of the carotid artery).

Coronary angioplasty

A surgical endovascular procedure performed to widen the narrowed (stenotic) coronary arteries of the heart.

Adventitial fibroblasts

Fibroblasts that are part of the area between the external elastic lamina and the outermost edge of a blood vessel (adventitia).

Lysyl oxidase

(LOX). An enzyme that promotes collagen and elastin crosslinking.

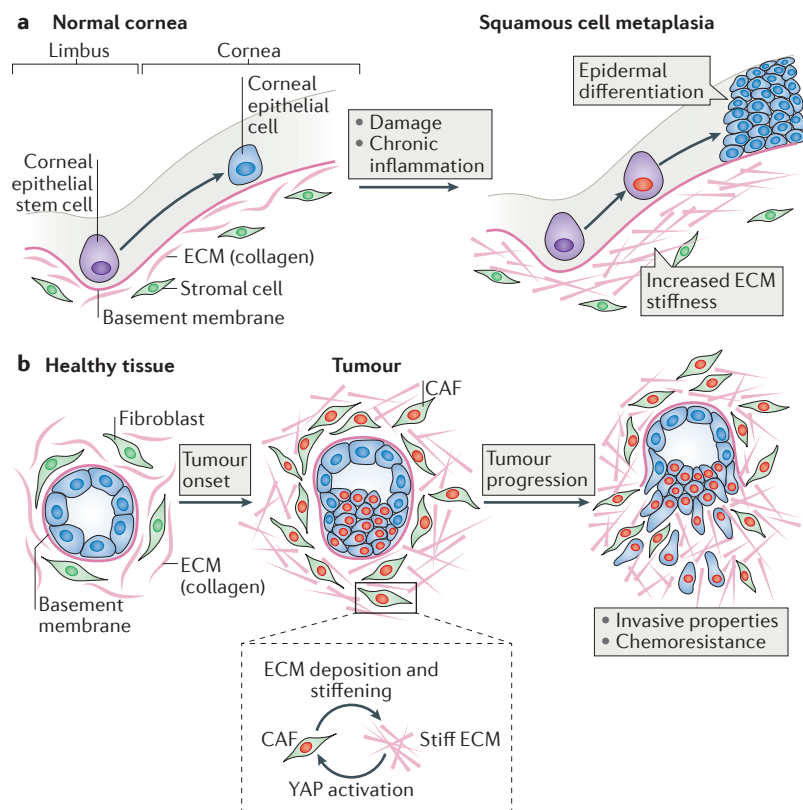


Figure 6 | Deregulation of YAP and TAZ mechanotransduction in disease. Aberrant mechanotransduction can lead to abnormal YAP and TAZ activation (red nuclei) in various pathological conditions. **a** | The change of fate of corneal epithelial cells into epidermal cells results in squamous cell metaplasia¹¹⁵, which is induced by tissue damage and chronic inflammation, resulting in extracellular matrix (ECM) stiffening. **b** | Cancer development is associated with the activation of YAP in cancer-associated fibroblasts (CAFs) by ECM stiffening^{36,66,117}; YAP promotes the activity of CAFs and leads to collagen deposition and further stiffening of the ECM. As a result, a mechanically activated positive-feedback loop operates, exacerbating the pathological outcome (see inset)³⁶. Stiff ECM also promotes YAP-mediated and TAZ-mediated mechanotransduction in the cells of the developing tumour, which promotes their metastatic dissemination and acquisition of chemoresistance. Intriguingly, a similar mechanical feedback mechanism is likely to also occur in pulmonary hypertension^{105,106} and fibrosis^{110,111}.

Hepatic stellate cells

Pericytes found in the perisinusoidal space of the liver; these are the major cell type involved in fibrosis in response to liver damage.

Metaplasia

Reversible transition of a differentiated cell type to another differentiated cell type that can be caused in a tissue by an abnormal stimulus; once the stimulus ceases, the tissue returns to its normal differentiation condition. Metaplasia is often considered an early phase of the tumorigenic process.

deaths worldwide. The cardinal traits shared by these diseases are the abnormal remodelling and excessive deposition of ECM by activated fibroblasts, also known as myofibroblasts, over a long period of time; this triggers tissue stiffening, cellular dysfunction and eventually organ failure. Thus, there is great interest in developing means to oppose or even reverse fibrosis. Although the cellular players involved are well understood, the signalling pathways underlying fibrosis are less clear¹⁰⁷. Importantly, YAP and TAZ mechanotransduction is emerging as a key player in these processes.

In liver fibrosis, YAP is activated in hepatic stellate cells in response to liver damage *in vivo* and to ECM stiffening *ex vivo*^{108,109}. Activation of hepatic stellate cells by tissue damage triggers YAP nuclear translocation and increases in YAP total levels and activity. This also results in increases in collagen deposition and smooth muscle actin expression, which are associated with the generation of myofibroblasts (FIG. 2). Interestingly, in the fibrotic areas of livers from patients with hepatitis C,

myofibroblasts displayed strong YAP nuclear staining, and hepatic stellate cell activation could be reversed by pharmacological inhibition of YAP¹⁰⁸.

YAP and TAZ are also induced in the lungs of patients with idiopathic pulmonary fibrosis, and YAP and TAZ sustain a pro-fibrotic transcriptional programme in human lung fibroblasts both *in vitro* and *in vivo*^{110,111} (FIG. 2), including increased collagen and fibronectin deposition and secretion of pro-fibrotic cytokines. Consistently, *Taz*-heterozygous mice were resistant to bleomycin-induced lung fibrosis¹¹². Of note, YAP-dependent and TAZ-dependent collagen deposition is itself fostered by ECM stiffness, indicating the presence of mechanically self-reinforcing positive loops, similar to the feedback regulation described for pulmonary hypertension.

A fascinating open issue is whether targeting YAP and TAZ mechanotransduction might have therapeutic effects or might even reverse tissue fibrosis.

YAP and TAZ as mediators of inflammatory responses.

Inflammation is the response to injury, which entails recruitment of leukocytes and plasma proteins, activation of a fibrogenic response and ECM remodelling. Acute inflammation is a transient response, integral to the regenerative response of normal tissues that stops after the healing process is completed. Importantly, YAP mediates epithelial tissue regeneration after inflammatory damage^{66,113}; in these contexts, YAP activation is Hippo/LATS independent but dependent on SRC-family kinases³³. SRC activity is a potent inducer of cytoskeletal remodelling downstream of FAK and integrin signalling^{59,114}, raising the enticing possibility that inflammation may activate YAP and TAZ also by inducing increased mechanoresponsiveness in epithelial cells.

The potential links between inflammation and YAP and TAZ mechanotransduction are particularly appealing in the context of chronic inflammatory diseases, which, differently from acute inflammation, represent persistent conditions, characterized by constant tissue damage and repair that progressively erode tissue integrity and function. The causes of chronic inflammation are poorly understood, but the persistence of a disturbed tissue architecture that never heals may causally contribute to such maladaptive responses. Although still awaiting further validation, a connection that links chronic inflammation, ECM stiffening and altered YAP and TAZ signalling has recently been demonstrated. In a mouse model of corneal squamous cell metaplasia, corneal wounding induces chronic inflammation and excessive deposition of ECM by the corneal stroma, followed by YAP and TAZ nuclear localization. In this mechanically aberrant environment, epithelial stem and progenitor cells are then driven to transdifferentiate into keratinizing, epidermal-like cells, resulting in metaplasia and ocular surface blindness¹¹⁵ (FIG. 6a).

Muscular dystrophy. Defects in YAP-mediated mechanosensing have been reported in patients carrying mutations in lamin A (LMNA) associated with congenital muscular dystrophy. These cells present several

defects in the organization of the cytoskeleton, causing aberrant YAP and TAZ mechanotransduction: YAP is cytoplasmic in healthy control myoblasts when these are cultured on a soft ECM, but it remains nuclear in soft hydrogel cultures of LMNA-mutant cells. Paradoxically, YAP activity is blunted in these mutant cells when they are challenged by cycles of stretch and relaxation¹¹⁶.

Instructive roles of YAP and TAZ in cancer development.

YAP and TAZ are pervasively activated in human cancers, where they are required to instruct malignant properties, including unrestrained proliferation, cell survival, chemoresistance and metastasis^{66,117}. These effects may be explained, at least in part, by the ability of active YAP and TAZ to convert benign neoplastic cells into cancer stem cells^{66,118}. It should be noted, however, that in spite of their widespread activation in tumours, activating mutations in *YAP* or *TAZ* have not been reported in human cancers; moreover, Hippo pathway mutations are rare in human tumours and not yet detected in carcinomas. Mechanical inputs from the aberrant tumour microenvironment are therefore prime candidates to induce YAP and TAZ overactivity in cancer cells. These inputs include aberrant tissue organization, accumulation of stromal cells, inflammation, increased compression forces and interstitial pressure, metalloproteinase-mediated ECM remodelling by cancer-associated fibroblasts (CAFs) and overall ECM stiffening^{1,119,120} (FIG. 6b).

YAP and TAZ regulation by the cytoskeleton might represent an interesting target for the design of anti-cancer therapeutics. Of note, recent studies suggest that mechanical activation of YAP and TAZ in cancer cells is also relevant for acquisition of chemoresistance^{121–123} (see also REF. 66 for review). However, given the toxicity of anti-cytoskeletal treatments, therapeutic options are currently limited to inhibitors of RHO and its upstream inducers, such as SRC, FAK and integrins. As in the case of atherosclerosis, drugs that target geranylgeranyl synthesis, thereby blocking RHO GTPase recruitment to the membrane, such as statins, bisphosphonates and geranylgeranyl transferase 1 inhibitors, display YAP and TAZ inhibiting abilities in several cancer cell types³². Statins and bisphosphonates also reduce the growth of tumour xenografts^{32,35,124}, although their effects appeared stronger *in vitro* than *in vivo*. Some epidemiological studies showed that patients under statin therapies display reduced risk to develop cancer¹²⁵, although there are conflicting data on this topic¹²⁶, highlighting the need for further clinical studies on the potential of these drugs for cancer treatment and/or prevention.

In several tumours, CAFs are responsible for increasing ECM stiffness and for changing the cellular composition of tumour niches by secreting chemokines, thereby promoting tumour invasion and angiogenesis. These properties rely on activation of YAP in CAFs and occur downstream of ECM stiffening³⁶. In turn, increased YAP activity in CAFs promotes contractility and restructuring of the ECM, locking fibroblasts in a vicious mechanically activated positive-feedback loop that self-sustains YAP and TAZ activation in both CAFs and tumour cells (FIG. 6b).

Perspectives and key questions

In this Review, we highlighted the role of YAP and TAZ as a molecular entry point to understand how mechanical forces impact cell behaviour and tissue pathophysiology from the micrometre scale to the macroscopic scale. However, despite rapid progress, this field is still in its infancy and several outstanding questions remain unanswered.

First, the mechanisms by which cell mechanics regulate YAP and TAZ remain elusive. F-actin integrity and organization is now broadly accepted as a dominant input to regulate YAP and TAZ. But how does it occur exactly? Many hypothetical scenarios can be proposed; for example, YAP and TAZ may be normally kept inactive by inhibitory proteins, and F-actin itself or F-actin-associated proteins may oppose this inhibitory association. Also unclear is where YAP and TAZ regulation by mechanotransduction occurs. Several lines of evidence indicate that nuclear localization of YAP and TAZ may not automatically translate into transcriptional activation. Actomyosin-generated cytoskeletal tension regulates nuclear shape and, intriguingly, force transfer from the cytoplasm to the nucleus through the linker of nucleoskeleton and cytoskeleton (LINC) complex is essential for activation of the YAP and TAZ pathway¹²⁷. Moreover, the nuclear lamina has been recently shown to be mechanosensitive and to affect YAP and TAZ activity¹²⁸. This raises the interesting question of whether nuclear mechanics are critical for YAP and TAZ activity. Unfortunately, the nuclear lamina and cytoskeleton are not preserved in a typical cell lysate, hampering our ability to capture perhaps the most relevant YAP and TAZ interacting partners by mass spectrometry.

The exact connections between Hippo signalling and mechanotransduction of YAP and TAZ are also elusive. Redundant lines of evidence from various model systems indicate that YAP and TAZ mechanotransduction occurs independently from Hippo kinases^{24–35}, although these two pillars of YAP and TAZ regulation must obviously engage in crosstalk to act coherently. There is currently no evidence that Hippo/LATS activity is patterned in contexts in which YAP and TAZ activity is regulated in an exquisitely cell-specific and temporal-specific manner⁶⁶. RHO and the F-actin cytoskeleton are overarching determinants for the effect of the Hippo kinases on YAP and TAZ, and the integrity of the F-actin cytoskeleton is essential for YAP inhibition by LATS. At the same time, it is also clear that LATS activity itself is under cytoskeletal control^{47,129} and that LATS can also affect the cytoskeleton²³. Addressing these issues is imperative to attain an integrated molecular overview of YAP and TAZ regulation by cell mechanics. This should shed light on one of the outstanding problems in cell biology, that is, understanding the nature of self-organizing properties of stem cells and tissues, whereby a sequence of mechanical and architectural checkpoints must be achieved to allow unperturbed development, tissue repair and homeostasis.

The role of YAP and TAZ and their regulation by the multitude of physical tissue deformations occurring during gastrulation and morphogenesis of distinct organs remain a largely unexplored area of investigation.

Myoblasts

Embryonic progenitor cells that can differentiate into muscle cells. Myoblasts that do not differentiate into muscle cells in the skeletal muscle fibre can turn into satellite cells (somatic muscle stem cells).

Linker of nucleoskeleton and cytoskeleton (LINC) complex

A protein complex associated with the nuclear membrane and composed of SUN domain and KASH domain proteins. This complex serves as a linker connecting nuclear lamins and chromatin with cytosolic actin microfilaments, microtubule filaments and intermediate filaments.

Evo-devo

(evolutionary developmental biology). A field in biological studies that compares the developmental processes of different organisms to trace their evolutionary relationships and to infer the evolution of developmental processes.

Embryonic overactivation of YAP and TAZ has been connected to organ overgrowth at least in some tissues (the liver, heart and stomach); intriguingly, this is a reversible process, as the organ shrinks back to its 'correct' size after normalization of YAP and TAZ levels. Thus, each cell of an organ must perceive the presence of all other cells in order to stop its growth when the organ has reached its final size. Diffusion of morphogen gradients is poorly suited to carry out these effects. Instead, mechanical forces transmitted through the interconnected network of cytoskeletal and ECM proteins may ideally serve as long-range messengers of this global control, informing individual cells with nanometre accuracy. This remains an enticing speculation, and in its favour, growth and shaping of the fly wing primordium is YAP dependent and controlled by global patterns of mechanical stress that originate from its stretched tissue border¹³⁰. As early as 1917, D'Arcy Thompson published his visionary book *On Growth and Form* (REF. 131), in which he proposed that "diagrams of forces" underlie tissue patterning and that force-oriented changes in the 3D shapes of tissues may dictate the evolutionary aspects of morphological diversities between related species. The integration of YAP and TAZ biology with evo-devo approaches, along with materials science and mathematical modelling, may offer fresh experimental perspectives to these visionary ideas. Crucially, addressing these issues would require the implementation of new tools allowing, on the one hand, the ablation of YAP and TAZ in individual organs during development and, on the other hand, genetic manipulation of cell mechanics *in vivo*.

One of the most remarkable properties of mechanically activated YAP and TAZ, at least *in vitro*, is their ability to trigger conversion of differentiated cells into their corresponding tissue-specific stem cells⁶¹. Is this exploited *in vivo*? Are YAP and TAZ specifically induced when the homeostatic, repressive tissue architecture is lost after damage and inflammation? Experimental overexpression of YAP also awakens regenerative potentials that are normally not exploited by mature tissues. Heart regeneration is a case in point, as forced YAP activation rescues cardiomyocyte proliferation and fosters organ recovery after myocardial infarction in mice^{99,100}. Is lack of regeneration linked to the establishment of a mechanical environment that is not permissive for proper YAP and TAZ activation? Can biomaterials be exploited as therapeutic tools to foster such latent healing powers?

Finally, what are the evolutionary roots of the YAP and TAZ involvement in mechanotransduction? The widespread relevance of a unique set of transcription factors, in multiple tissues and cell types, downstream of a broad array of pathophysiological mechanical stresses, is both stunning and surprising. Of note, *D. melanogaster* YAP (Yki) is also under the control of F-actin organization^{37,49}. All this probably reflects a conserved function of YAP and TAZ in extracting information from mechanical signals. Intriguingly, the appearance of YAP predates multicellularity, being first detected in the genome of the closest unicellular relatives of the animals¹³². It would be interesting to test whether YAP activity is connected to sensing substrate mechanics in these unicellular systems as well.

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Author contributions

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