IMMUNOLOGY

Mechanosensation to inflammation: Roles for YAP/TAZ in innate immune cells

Vijaykumar S. Meli[1,2]†, Praveen Krishna Veerasubramanian[1,2]†, Timothy L. Downing[1,2,3,4], Wenqi Wang[5], Wendy F. Liu[1,2,6,7,8]*

Innate immune cells are responsible for eliminating foreign infectious agents and cellular debris, and their ability to perceive, respond to, and integrate biochemical and mechanical cues from their microenvironment eventually determines their behavior. In response to tissue injury, pathogen invasion, or a biomaterial implant, immune cells activate many pathways to initiate inflammation in the tissue. In addition to common inflammatory pathways, studies have demonstrated the role of the mechanosensitive proteins and transcriptional coactivators YAP and TAZ (YAP/TAZ) in inflammation and immunity. We review our knowledge of YAP/TAZ in controlling inflammation and immunity in innate immune cells. Furthermore, we discuss the roles of YAP/TAZ in inflammatory diseases, wound healing, and tissue regeneration and how they integrate mechanical cues with biochemical signaling during disease progression. Last, we comment on possible approaches that can be exploited to harness the therapeutic potential of YAP/TAZ in inflammatory diseases.

INTRODUCTION

Cells in living organisms experience many biophysical and biochemical cues from the surrounding cells and the extracellular matrix (ECM), which together influence their behavior. To proliferate, differentiate, and regenerate, cells must integrate and respond to these cues that emerge from their immediate milieu. Furthermore, dysregulated coordination between cells and their microenvironment drives multiple diseases, including atherosclerosis, fibrosis, and cancer (*1*, *2*). Over the years, whereas biochemical signals, such as cytokines, chemokines, and growth factors that govern various immune cell functions, have been relatively well studied, the contributions of biophysical cues have often been overlooked (*3*). Emerging evidence underpins the importance of mechanical signals as fundamental regulators of cell behavior (*1*, *4*); however, how these mechanical cues are perceived and relayed at the molecular level to regulate gene expression and what their distinct roles are in immune cells still remain elusive. The two related and conserved transcriptional regulators, Yes-associated protein (YAP; which is encoded by *YAP1*) and its paralog transcriptional coactivator with PDZ-binding motif (TAZ; which is encoded by *WWTR1*) (together referred to as YAP/TAZ) are involved in sensing the microenvironmental landscape and modulating cell function (*1*). These transcriptional coactivators were primarily identified as gene products that promote cell proliferation, differentiation, and survival from stress upon nuclear translocation (*5*). However, studies showed that they also have an essential role in innate immunity and inflammatory diseases (*6*–*14*). Here, we describe our current understanding of YAP/TAZ functions in innate immune cells in response to biomechanical perturbations during physiological and pathological conditions.

The Hippo pathway is a kinase cascade that was first identified in *Drosophila* and is highly conserved in mammals. This pathway controls tissue growth and organ size by suppressing cell proliferation and promoting cell death and is therefore often referred to as a tumor-suppressor pathway (*1*). The core Hippo pathway consists of the upstream kinase Hippo [mammalian STE20-like serine/threonine kinases (MST1/2) in mammals], which phosphorylates and activates the downstream kinase Warts (Wts) [large tumor suppressor kinase (LATS1/2) in mammals]. Wts then phosphorylates the transcriptional coactivator Yorkie (Yki) (YAP/TAZ in mammals) to inhibit its nuclear translocation. When the Hippo pathway is active, this inhibitory phosphorylation by LATS1/2 leads to the cytoplasmic retention of YAP/TAZ by 14-3-3 proteins, resulting in proteasomal degradation (*15*). On the other hand, when the Hippo pathway is inactive, YAP/TAZ accumulate in the nucleus and bind to the transcription factors TEA domain–containing transcription factors 1 to 4 (TEAD 1 to 4), which are orthologs of *Drosophila* Scalloped, to transcribe many genes involved in cell proliferation and survival (*2*, *16*, *17*). The canonical Hippo signaling pathway is thought to primarily inhibit YAP/TAZ activation, suppressing cell proliferation and promoting apoptosis during tissue homeostasis (Fig. 1) (*18*). However, studies have revealed additional kinases and posttranslational modifications independent of the canonical Hippo pathway that can also regulate YAP/TAZ (*9*, *19*–*21*). Thus, YAP/TAZ are regulated by Hippo-dependent and Hippo-independent pathways, which together determine their subcellular localization and stability and their effects on cell behavior.

The role of YAP/TAZ in sensing mechanical cues has been studied extensively across different cell types and tissues. Although immune cells function within dynamic mechanical niches and experience varied stiffness, shear flow, and dynamic cell-cell interactions, their general nonadherent nature and inherent structural plasticity have led them to be relatively understudied in the

[1]Department of Biomedical Engineering, University of California, Irvine, Irvine, CA 92697, USA. [2]UCI Edwards Lifesciences Foundation Cardiovascular Innovation and Research Center (CIRC), University of California, Irvine, Irvine, CA 92697, USA. [3]NSF-Simons Center for Multiscale Cell Fate Research, University of California, Irvine, Irvine, CA 92697, USA. [4]Department of Microbiology and Molecular Genetics, University of California, Irvine, Irvine, CA 92697, USA. [5]Department of Developmental and Cell Biology, University of California, Irvine, Irvine, CA 92697, USA. [6]Department of Chemical and Biomolecular Engineering, University of California, Irvine, Irvine, CA 92697, USA. [7]Department of Molecular Biology and Biochemistry, University of California, Irvine, Irvine, CA 92697, USA. [8]Institute for Immunology, University of California, Irvine, Irvine, CA 92697, USA.
*Corresponding author. Email: wendy.liu@uci.edu
†These authors contributed equally to this work.

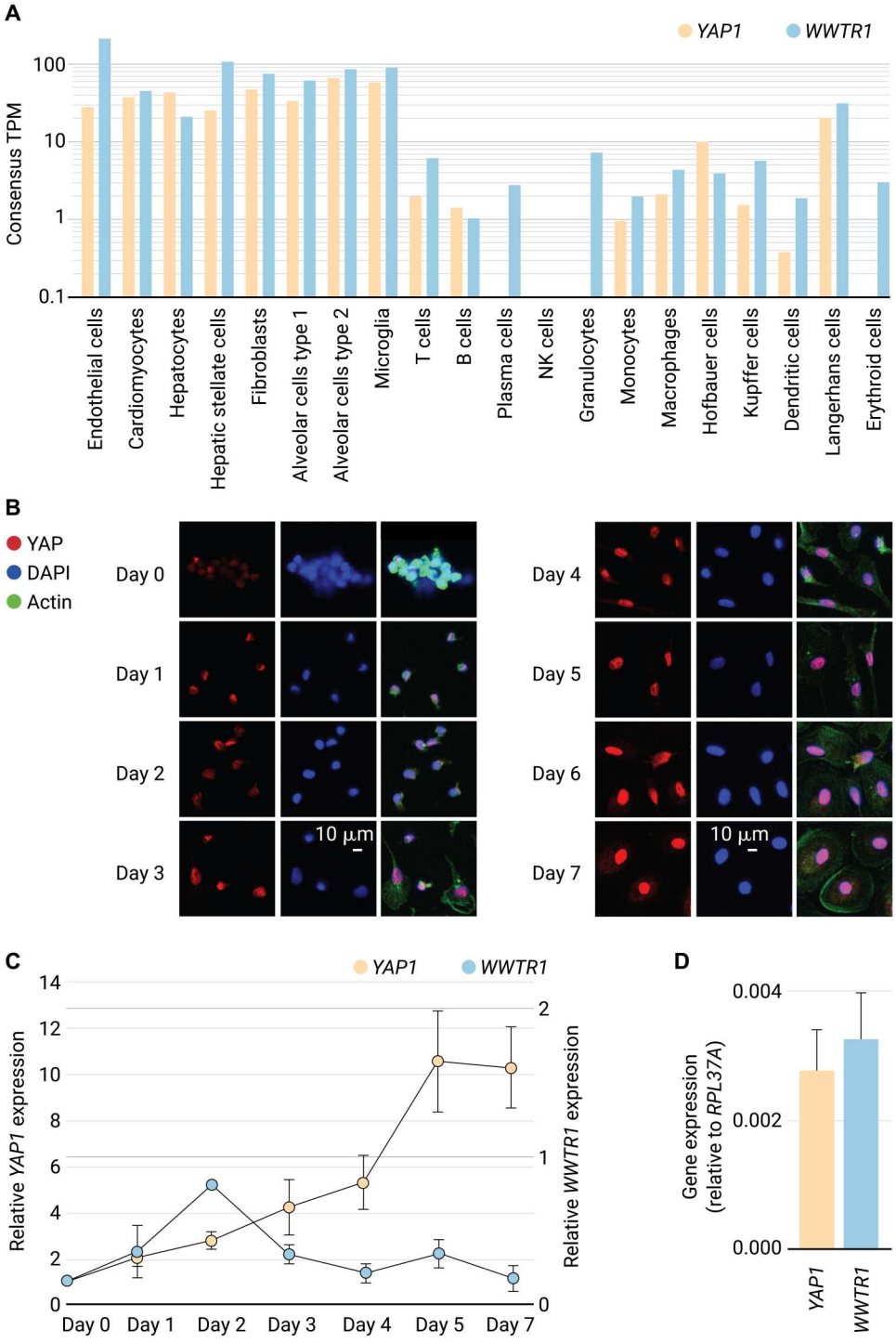

Fig. 1. YAP abundance, but not that of TAZ, increases during monocyte-macrophage differentiation. (**A**) Data from the Human Protein Atlas (*25*) show relative YAP (*YAP1*) and TAZ (*WWTR1*) expression in the indicated cells. TPM, transcripts per million. (**B**) Fluorescence microscopy of YAP during the differentiation of monocytes (from PBMCs) to macrophages over time. (**C**) The expression of *YAP1* but not *WWTR1* increases over time as monocytes differentiate into macrophages. (**D**) *YAP1* and *WWTR1* are expressed in macrophages. Data in (B) to (D) are adapted from the work of Meli *et al.* (*11*) with permission. DAPI, 4′,6-diamidino-2-phenylindole.

context of mechanobiology (*21*). Early studies found that YAP is not detected in peripheral blood leukocytes (*22*), and its abundance is low in various immune cells, including THP-1 monocytes and peritoneal macrophages, as compared with that in human embryonic kidney (HEK) 293 and mouse embryonic fibroblasts (*23*, *24*), questioning its importance and functionality in immune cells. *YAP1* expression in monocytes and macrophages is up to 10-fold less than that in nonimmune cells, such as fibroblasts, endothelial cells, and epithelial cells (Fig. 1A) (*25*). However, we showed that *YAP1*, but not *WWTR1*, expression increases upon differentiation of monocytes to adherent macrophages (Fig. 1, B to D) (*11*). Moreover, other studies have demonstrated context-dependent YAP/TAZ functions in immune cells, controlling inflammation and immunity (*11*). Hippo signaling and the innate immune system cross-talk with each other, resulting in antiviral and antibacterial activity. In addition, YAP/TAZ are abnormally regulated in many inflammatory diseases, during wound healing, and during tissue regeneration (*11*, *13*). Despite these insights, the importance of cues from the mechanical niche in which innate immune cells reside and differentiate is still poorly understood.

Mechanical cues control YAP/TAZ activity through both Hippo-dependent and Hippo-independent pathways. Cells perceive ECM stiffness and shear forces through integrins, the engagement and activation of which promote focal adhesion assembly. ECM rigidity is relayed by the Ras-related guanosine triphosphatase (GTPase) RAP2, which is activated under conditions of low stiffness and binds to mitogen-activated protein kinases (MAPKs) and Rho GTPase–activating protein 29, activating LATS1/2 and inhibiting YAP/TAZ (*26*). ECM rigidity induces Rho-associated coiled-coil kinase (Rho-ROCK) signaling, causing F-actin polymerization and actomyosin contractility, leading to the nuclear translocation of YAP/TAZ (*27*, *28*). In response to mechanical stimuli, the cytoskeleton remodels, leading to changes in F-actin abundance, which can modulate YAP/TAZ activity (*29*–*33*). Disassembly of F-actin activates LATS1/2 and promotes YAP phosphorylation. In addition, MAPKs, protein kinase A, and thousand and one kinases can activate LATS1/2, but their regulation by F-actin remains unclear (*27*, *30*, *34*–*36*). Furthermore, angiomotins (AMOTs) bind to F-actin and inhibit Hippo signaling (*37*). When F-actin abundance is low, AMOT binds to MST1/2 and LATS1/2 and activates these kinases to sequester YAP in the cytoplasm (*38*, *39*). Neurofibromatosis 2 (NF2) is controlled by mechanical stimuli and AMOTs, and it also regulates the Hippo pathway. AMOT binds to NF2 to activate LATS1/2 or binds directly to YAP in a Hippo-independent manner and inhibits its nuclear translocation (*40*). However, F-actin assembly caused by mechanical stimuli promotes the binding of AMOT to actin, rendering MST1 and LATS1/2 inactive and therefore facilitating the nuclear translocation of YAP. In addition, signaling components of focal adhesions, including focal adhesion kinase and Src, are required for YAP/TAZ activity, and Src phosphorylates YAP directly (*41*, *42*) or activates YAP indirectly by phosphorylating LATS1 (*43*). However, it is still unclear whether direct phosphorylation of YAP/TAZ by Src affects YAP/TAZ activity directly or indirectly through the cytoskeleton. In addition, the co-receptor for the inflammatory cytokine interleukin-6 (IL-6), gp130, associates with Src, leading to the phosphorylation of YAP and promoting its nuclear translocation in epithelial cells (Fig. 2) (*44*). Src also promotes inflammation in macrophages (*45*), but its regulation of YAP in these cells has not yet been demonstrated.

In addition to forces at focal adhesions, tension sensing at the adherens junction component of cell-cell contacts can regulate YAP/TAZ. Under conditions of high tension (low cell density), α-catenin and β-catenin bind to actin stress fibers, *Lin*-11 *Isl*-1 *Mec*-3–containing 1 (LIMD1), and vinculin proteins. LIMD1 promotes the recruitment of LATS1/2 to the junctions, and vinculin recruits thyroid hormone receptor interactor 6 (TRIP6), which inhibits LATS1/2 activation, leading to the nuclear translocation of YAP/TAZ (*27*). On the other hand, high cell densities (reduced tension) lead to the loss of actin stress fibers, which inhibits the recruitment of LIMD1, vinculin, TRIP6, and LATS1/2 to the junction to suppress YAP/TAZ nuclear translocation (*27*). In addition, contraction of the circumferential actin belt underlying the adherens junctions due to high cell density in certain cell types can regulate YAP/TAZ activity independently of the Hippo pathway (*46*). Mechanistically, high cell density results in the dissociation of NF2 from adherens junctions so that it binds to YAP/TAZ to suppress YAP/TAZ nuclear localization. Nonetheless, whereas upstream regulators of YAP/TAZ in response to mechanical stimuli are better characterized in other cell types in comparison to immune cells, it is possible that upstream regulators in immune cells may function similarly to those in other cell types.

Here, we review the current understanding of YAP/TAZ in innate immune cell function, inflammation, and immunity. We discuss the potential roles of biophysical cues that influence YAP/TAZ in innate immune cells. We then describe the role of YAP/TAZ in inflammatory diseases, wound healing, and tissue regeneration. We propose a physical basis for these disease conditions caused by changes in tissue structure and remodeling, which might contribute to perturbed mechanochemical signaling. Last, we comment on the therapeutic potential of controlling and potentially co-opting YAP/TAZ in inflammatory diseases.

YAP/TAZ REGULATE IMMUNE CELL FUNCTION AND INFLAMMATION

YAP/TAZ in the differentiation and polarization of innate immune cells

It was previously postulated that Hippo signaling might be involved in the regulation of hematopoietic stem cells that give rise to myeloid and lymphoid cell lineages, given its known role in the proliferation of undifferentiated progenitor cells, stem cell self-renewal and differentiation, and organ size in various tissues (*47*–*49*). However, ectopic expression of YAP does not influence hematopoietic stem cell function under physiological or hematopoietic stress conditions (*50*). YAP regulates the differentiation and activity of osteoclasts, specialized cells derived from the monocyte-macrophage hematopoietic lineage that are formed at or near the bone surface (*51*, *52*). Zhao *et al.* (*51*) demonstrated that short hairpin RNA (shRNA)–mediated knockdown of YAP in bone marrow–derived macrophages (BMDMs) and inhibiting the YAP-TEAD association with the small molecule verteporfin prevent the formation of multinucleated osteoclasts. Despite the relatively low abundance of YAP in leukocytes (*22*), our group and others showed that YAP plays critical roles in macrophage function. YAP abundance appears to increase not only with the differentiation of peripheral blood mononuclear cell (PBMC)–derived monocytes to macrophages (*11*) but also in macrophages undergoing proinflammatory (*11*) and reparative programs (*12*). In dendritic cells, the kinases

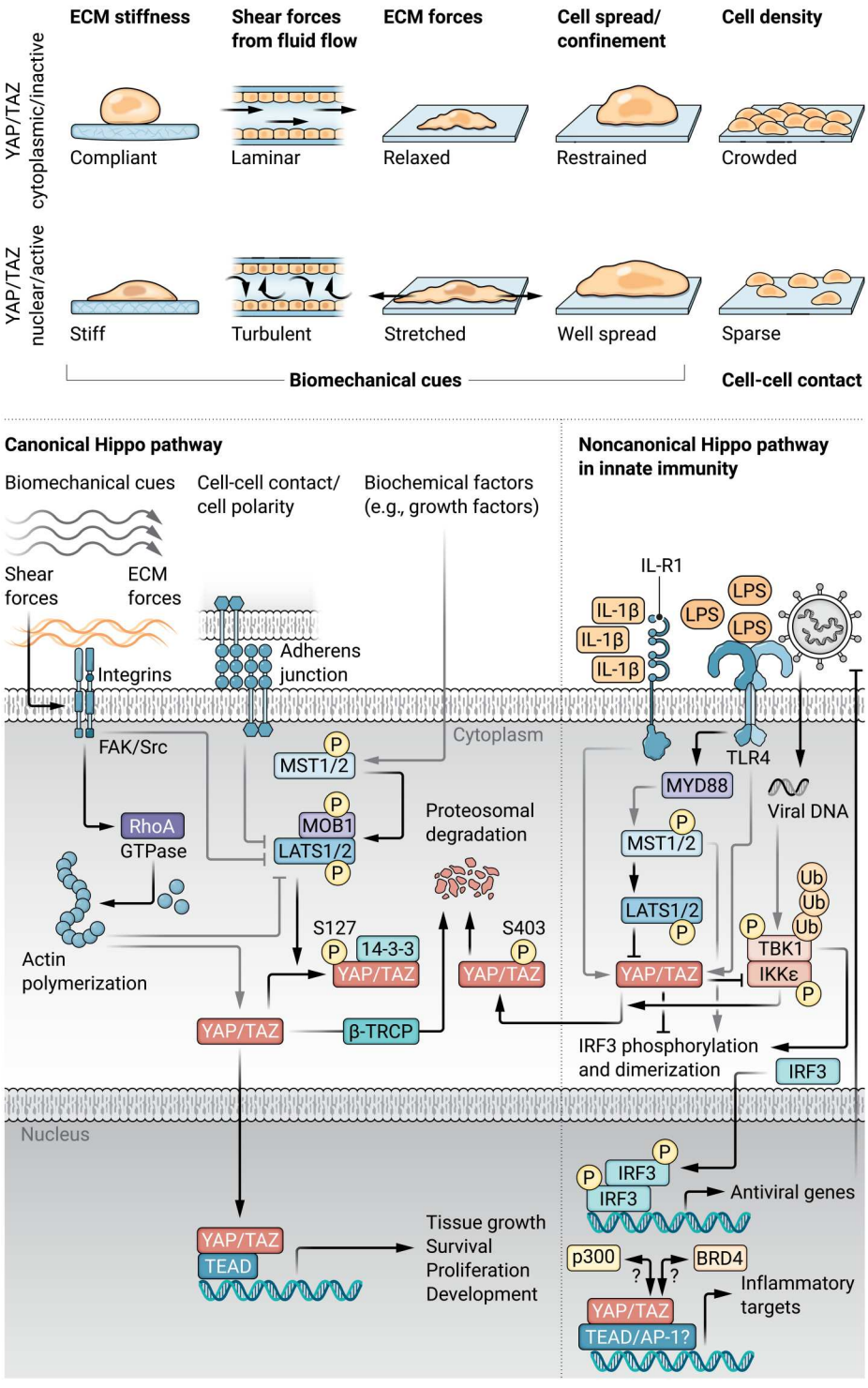

Fig. 2. The canonical and noncanonical Hippo pathways modulate innate immunity and inflammation. (**Top**) YAP/TAZ regulation is mechanically controlled through biophysical cues such as stiffness, shear flow, ECM forces, and cell density. (**Bottom**) Canonical regulation of YAP/TAZ occurs through upstream Hippo components and regulators that integrate biochemical and biomechanical cues to exert locational control over YAP/TAZ. The transcriptional activity of YAP/TAZ in the nucleus results in increased tissue growth, survival, proliferation, and development. In innate immune cells, YAP/TAZ localization is also influenced by bacterial and viral response pathways. In addition, cytoplasmic YAP/TAZ suppresses activation of the antiviral transcription factor IRF3. Solid arrows indicate evidence for direct effects, whereas dashed arrows indicate evidence for indirect (cascading) effects.

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MST1/2 selectively drive antigen presentation to $CD8^+$ T cells by integrating metabolic activity and cytokine signaling (*53*). These examples highlight the interplay between Hippo signaling and innate immune cell differentiation, signaling, metabolic reprogramming, and phenotype-specific function.

The few studies examining the role of YAP/TAZ in macrophage polarization have yielded somewhat varied results. In one study, YAP promoted the polarization of peritoneal macrophages (PEMs) and BMDMs into cells of the classical inflammatory M1 phenotype, whereas YAP deficiency enhanced their differentiation into the pro-healing, alternatively activated M2 subset, suggesting that YAP regulates the balance between M1 and M2 macrophage differentiation (*13*). However, in another study, the YAP inhibitor verteporfin attenuated the M2 polarization of RAW 264.7 cells (*54*). Similarly, TAZ ablation reduced the polarization of M2 macrophages in a kidney fibrosis model (*54*). Together, these findings suggest that the role of YAP/TAZ in the differentiation and polarization of immune cells is still emerging and that more work is warranted to define the specific roles of YAP/TAZ in hematopoietic stem cell lineage determination, the differentiation of myeloid cells, and the polarization and function of macrophages. Considering that most studies examining the role of YAP/TAZ in immune cell function have been performed with cells on standard tissue culture plastic substrates, it is likely that the mechanical signals presented by the cell culture environment influenced the experimental outcomes. Furthermore, observed discrepancies in YAP/TAZ abundance between cells may be attributed to different tissue or cell sources as well as the differentiation methods used to obtain macrophages.

The Hippo pathway in inflammation and immunity

Innate immune cells express pattern recognition receptors (PRRs) as a surveillance system to respond to pathogen-associated

Table 1. Effects of inflammatory biochemical stimuli on YAP/TAZ abundance. HUVECs, human umbilical vein endothelial cells; qRT-PCR, quantitative reverse transcription polymerase chain reaction.

	Biochemical stimulus	Findings	Reference
Cells			
HUVECs	TNF-α (20 ng/ml)	Increased YAP nuclear localization (Western blotting and immunofluorescence)	(*127*)
HUVECs and human lung microvascular endothelial cells	LPS (1 μg/ml), TNF-α (10 ng/ml), 500 μM H_2O_2	Increased YAP abundance (Western blotting)	(*128*)
HEK 293 cells	TNF-α (10 ng/ml)	Increased YAP/TAZ abundance and nuclear localization in high-density cell cultures (Western blotting and immunofluorescence)	(*129*)
Macrophages derived from human PBMCs	LPS (10 ng/ml)	Increased YAP nuclear translocation	(*11*)
Murine peritoneal macrophages and BMDMs	IL-1β (10 ng/ml)	Increased YAP abundance and nuclear translocation (Western blotting)	(*9*)
Murine KCs and RAW264.7 cells	LPS (1 μg/ml)	Increased YAP abundance (immunofluorescence and qRT-PCR)	(*14*)
Murine peritoneal macrophages	LPS and IFN-γ	Increased YAP abundance and nuclear translocation (Western blotting and immunofluorescence)	(*13*)
Murine peritoneal macrophages and BMDMs	LPS (100 ng/ml) and IFN-γ (10 ng/ml; costimulation)	Increased YAP/TAZ abundance (Western blotting)	(*12*)
Murine peritoneal macrophages and THP-1 cells	IL-4 and IL-13 (10 ng/ml each; costimulation)	Reduced YAP abundance (Western blotting)	(*13*)
Tissues, animal models, or humans			
Murine lung tissue	LPS challenge	Increased YAP abundance in isolated lung endothelial cells and non-endothelial cells (Western blotting)	(*128*)
Murine liver tissue	LPS challenge	Increased YAP abundance in isolated KCs (flow cytometry and qRT-PCR)	(*14*)
Murine lung tissue	Mechanical ventilation–associated injury and inflammation	Increased YAP abundance (Western blotting and qRT-PCR)	(*10*)
Murine atherosclerotic tissue	Hypercholesteremia-induced atherosclerosis	Increased YAP/TAZ abundance in atherosclerotic carotid artery and aortic arch (immunofluorescence)	(*75*)
Murine and human atherosclerotic tissues	Hypercholesteremia-induced atherosclerosis	Increased YAP abundance in $CD68^+$ cells with disease progression (immunofluorescence and Western blotting)	(*9*)
Murine foam cells	Hypercholesteremia-induced atherosclerosis	Increased YAP abundance in $CD45^+$ $CD11b^+$ $F4/80^+$ cells with disease progression (flow cytometry)	(*9*)
Human patient–derived PBMCs	ST-segment elevated MI	Increased YAP abundance correlating with increased plasma IL-1β concentrations	(*9*)
Murine NASH liver	Hypercholesteremia-induced NASH	Increased YAP abundance in total tissue and $F4/80^+$ cells (Western blotting, flow cytometry, and immunofluorescence)	(*14*)

molecular patterns or host-derived, damage-associated molecular patterns. Signaling from PRRs results in the production of transcription factors that induce the expression of genes whose products are needed to eliminate host debris and foreign infectious agents and for the initiation of inflammation (*55*). Common inflammatory pathways stimulated in innate immune cells include those involving nuclear factor κB (NF-κB) subunits, MAPKs, and Janus kinase–signal transducer and activator of transcription proteins (*56*). However, evidence also suggests that the Hippo pathway is connected to innate and adaptive immune responses (*11*, *15*, *57*, *58*). One of the earliest pieces of evidence of the involvement of YAP in immunity came from studies of *Drosophila*, which showed that the YAP homolog Yki directly regulates the inhibitor of κB (IκB) homolog Cactus to control the antimicrobial response. Exogenous overexpression of Yki in fat bodies, the *Drosophila* immune organ, increases the abundance of *Cactus* transcripts and reduces the production of antimicrobial peptides, leading to susceptibility to Gram-positive bacteria (*59*). Upon infection by Gram-positive bacteria, signaling by Toll-like receptors (TLRs; a family of PRRs) causes the nuclear-to-cytoplasmic translocation of Yki, resulting in reduced production of Cactus and relieving the NF-κB family transcription factors Dorsal and Dorsal-related immune factor from its inhibitory effects (*59*).

The role of YAP in macrophage inflammation has been demonstrated. We showed that YAP/TAZ knockdown in human monocyte-derived macrophages substantially reduces the amount of tumor necrosis factor–α (TNF-α) secreted in response to the TLR4 agonist lipopolysaccharide (LPS) (*11*). Consistent with this observation, deletion of YAP in mouse myeloid cells leads to the suppression of LPS-induced systemic inflammation (*60*). In Kupffer cells (KCs) in the liver, LPS induces the accumulation of YAP and enhances hepatic inflammation and the production of proinflammatory cytokines, which is suppressed by knocking out YAP (*14*). YAP is mostly localized to the nucleus in cells cultured on stiffer substrates irrespective of the biochemical microenvironment, although some studies showed that the inflammatory cytokine IL-1β and LPS (*11*) enhance the nuclear localization of YAP in macrophages cultured on tissue culture plastic. Various biochemical stimuli that cause inflammation are observed to have effects on YAP/TAZ abundance (Table 1). In addition, culturing cells on stiffer substrates enhances the production of inflammatory mediators, such as IL-1β, IL-6, TNF-α, and TLR4 (*61*), which also promote the nuclear localization of YAP, leading to positive feedback and increased inflammation. In contrast, when macrophages are cultured on soft substrates, YAP is mostly cytoplasmic, and the stimulation of cells with LPS does not enhance its nuclear translocation (*11*); however, overexpressing the phosphorylation-resistant YAP mutant YAP-5SA in cells cultured on a soft substrate substantially enhances LPS-induced inflammatory responses. Similarly, pharmacological inhibition of or deficiency in TAZ suppresses stiffness-induced inflammatory cytokine production by bone marrow–derived dendritic cells (*62*). Thus, nuclear YAP enhances the inflammatory activation of innate immune cells and the ability of the cells to sense mechanical cues to tune their functional outcome.

In addition to their roles in transcriptional regulation, YAP/TAZ also directly regulate innate immune responses in the cytoplasm, and the notion that YAP/TAZ are functional only in the nucleus is undergoing revision. Evidence of YAP/TAZ physically binding to transcription factors and kinases in the cytoplasm to regulate immune responses is emerging. YAP inhibits the antiviral immune response, and its deficiency in PEMs and BMDMs increases the antiviral response (*63*). Macrophages isolated from myeloid cell–specific, YAP-deficient mice show increased expression of interferon beta 1 (*Ifnb1*) and of downstream chemokine-encoding genes upon incubation with viruses or viral RNA mimics. Therefore, YAP deficiency in myeloid cells protects mice from viral infection. Mechanistically, YAP in the cytoplasm inhibits the dimerization of the transcription factor interferon regulatory factor 3 (IRF3) and its translocation to the nucleus. However, these in vitro experiments were performed with cells cultured on tissue culture plastic, which potentially could influence the findings. Independently of YAP, another upstream Hippo pathway kinase, MST1, suppresses antiviral immunity induced by cytosolic RNA and DNA by directly binding to and phosphorylating IRF3 (*64*). Similarly, cytoplasmic YAP/TAZ associate directly with and suppress the kinases TANK-binding kinase 1 (TBK1) and IκB kinase ε, thereby inhibiting the antiviral response. Furthermore, knockdown of YAP/TAZ can enhance the antiviral response (*24*). This study showed the role of Hippo signaling in determining nucleic acid sensing and innate antiviral immunity through YAP/TAZ-mediated TBK1 blockade. Considering that the classic functions of YAP/TAZ are related to cell survival (*2*) and sensing the mechanical environment, it will be interesting to better understand how the biophysical environment regulates responses to viral RNA and DNA. It is possible that the increased amount of phosphorylated cytoplasmic YAP is targeted for proteasomal degradation in cells in soft environments (*11*) and is thus unable to suppress TBK1 activation (*24*) or inhibit IRF3 dimerization (*63*), which would therefore enhance the antiviral response. On the contrary, cells in a soft environment might be sensitive to viral RNA and DNA because of the reduced amounts of phosphorylated TBK1 (*65*) and phosphorylated NF-κB (*11*), adding more complexity to the regulation of antiviral immunity by YAP and possibly TAZ.

Epigenetic and chromatin regulatory roles of YAP/TAZ

YAP/TAZ orchestrate transcriptional control by integrating epigenetic regulators in the nucleus to modulate chromatin accessibility. Whereas many studies exploring YAP/TAZ were performed in the context of tumorigenesis, not much is known about the epigenetic influences of YAP/TAZ on innate immunity and inflammation. Interestingly, several of the epigenetic enzymes that are implicated in tumorigenesis studies are also involved in inflammation and immunity. Here, we shed light on the associations between YAP/TAZ and inflammatory epigenetic proteins, such as the histone acetyltransferase (HAT) p300 and the histone acetylation reader bromodomain-containing protein 4 (BRD4), and we speculate on the importance of these associations. The inflammatory transcriptional activity of YAP/TAZ likely involves association with inflammatory transcriptional factors and epigenetic determinants such as BRD4 and p300.

Histone acetylation is generally associated with regions of chromatin that are "open" for transcription. Enhancers occupied by YAP/TAZ are usually acetylated at histone H3 lysine-27 (H3K27Ac), a modification that is facilitated by the HAT activity of p300 (*66*, *67*). Consistently, p300 is enriched at enhancers that are associated with YAP binding (*67*), and it is also physically associated with the YAP/TAZ-TEAD complex (*68*). The association of

YAP/TAZ with p300 is biomechanically influenced. Cell crowding, which leads to the cytoplasmic localization of YAP in some cells, also reduces the occupancy of p300 and H3K27Ac on YAP target gene enhancers in cancer cells (*67*). Furthermore, silencing YAP results in reduced p300 association and H3K27Ac marks on enhancers of target genes, illustrating potential nonredundant roles for YAP in p300 recruitment to enhancers (*67*). Thus, association with p300 confers YAP/TAZ with the ability to target histone acetylation toward distal regulatory elements and activate nearby genes. The nuclear localization signal (NLS) on p300 enhances TAZ nuclear import by associating with it in the cytoplasm (*69*). p300 is a proinflammatory HAT that cooperates with NF-κB to drive proinflammatory gene programs, and the genetic and pharmacological inhibition of p300 leads to the suppression of inflammatory transcription (*70*). Macrophages cultured on soft fibrin substrates have less histone H3 acetylation and reduced YAP nuclear translocation when compared with cells cultured on stiff substrates, and further studies are required to determine whether macrophage mechanosensation can modulate epigenetic modifications through YAP (*11*). Drawing from these reports, we posit that p300 is recruited by YAP/TAZ-containing complexes to enhance its nuclear import and remodel chromatin to enable inflammatory transcription in conjunction with proinflammatory transcriptional factors.

BRD4, an important histone acetylation reader (including of H3K27Ac marks) and epigenetic mediator of inflammation (*71*), associates with the YAP/TAZ-TEAD complex in cancer cells (*68*). BRD4 is a cofactor that is required for YAP/TAZ-mediated sustained oncogene transcription in cancer, as evidenced by the dynamics of YAP/TAZ and its engagement with BRD4 at promoters and distal enhancers. BRD4 is recruited to chromatin by YAP/TAZ in MDA-MB-231 cells (*68*). In the absence of YAP/TAZ, even overexpression of BRD4 does not result in the increased expression of YAP/TAZ target genes, including those involved in cell proliferation and survival (*68*). Whereas these findings highlight the interdependence between YAP/TAZ and BRD4 proteins in orchestrating the epigenetic regulation of cell proliferation and pro-cancer behaviors, a similar nexus in immune cells driving inflammation has not yet been shown. Given the prominent proinflammatory roles of BRD4 and YAP/TAZ in the nucleus, studies that explore this nexus from the context of innate immunity and inflammation are highly warranted.

Macrophage YAP/TAZ aggravate inflammation and contribute to inflammatory diseases

A large and growing body of studies has shown that pathological YAP/TAZ activation in cancer and inflammatory diseases promotes inflammation indirectly by stimulating the production of chemokines (such as CCL2 and CXCL1) and driving macrophage recruitment (*6*, *7*, *72*). Moreover, increased YAP/TAZ abundance or hyperactivation through greater nuclear translocation is a recurring event in such diseases, underscoring the possibility of inherent mechanochemical signaling (*8*, *11*, *14*). Congruently, other studies have implicated the transcriptional activity of YAP/TAZ in innate immune cells in the progression of inflammatory diseases. Here, we explore reports on direct inflammatory influences stemming from YAP/TAZ transcriptional activity in macrophages and underscore their relevance in inflammatory diseases besides cancer (Fig. 3). We also examine the roles of aberrant biomechanical cues that may potentiate inflammation through YAP/TAZ activation.

Atherosclerosis is a progressive vascular disease characterized by fatty plaque buildup in the intima, monocyte/macrophage recruitment, and chronic local inflammation. Atheroprone regions of vessels experience disturbed flow and oscillatory shear forces that elicit vascular inflammation and proliferation (*73*). In addition, it has been hypothesized that the increased macrophage infiltration and matrix degradation observed in the disease causes the loss of elastic laminae, resulting in the up to 100% thicker and 50% stiffer vascular walls that are observed in diseased aortas (*74*). En face analysis of atheroprone areas of murine endothelium revealed increased YAP/TAZ nuclear localization and target gene expression compared with atheroprotective areas (*75*). Accordingly, YAP and TAZ play mechanosensing roles in atherosclerosis.

YAP/TAZ in macrophages and endothelial cells are both essential in atherogenesis. In endothelial cells, YAP/TAZ nuclear translocation increases in response to disturbed flow, promoting increased proliferative and proinflammatory gene expression (*72*, *75*). Inhibition of YAP/TAZ in endothelial cells inhibits inflammation, leukocyte attachment, and infiltration and is atheroprotective in *Apoe*$^{-/-}$ mice fed an atherogenic, high-fat diet (HFD) (*72*, *75*, *76*). One study directly implicates macrophage YAP in inflammation associated with atherosclerosis (*11*). TNF receptor–associated factor 6, a component of IL-1β signaling, ubiquitylates YAP at Lys^{63}, which stabilizes YAP, causing its dissociation from AMOT, an interaction that otherwise sequesters YAP in the cytoplasm. This leads to increased YAP localization in the nucleus, where it induces proinflammatory gene expression, exacerbating atherosclerosis in the process (*11*). In addition, active YAP results in increased macrophage migration through its effects on the expression of genes encoding chemokines, which contributes to the enhanced monocyte-macrophage infiltration into the plaque observed in hypercholesteremic *Apoe*$^{-/-}$ mice whose macrophages overexpress YAP (*11*). Overall, these results highlight YAP as an attractive target to curb atherogenesis. Further studies are required to examine how synergistic biophysical cues, such as disturbed flow and vascular stiffening, regulate YAP/TAZ in macrophage-mediated inflammation in atherosclerosis.

Nonalcoholic steatohepatitis (NASH) is a critical liver disease that has its origins in obesity-induced fatty liver (steatosis). NASH manifests as liver inflammation, fibrosis, and hepatocellular damage that progresses to scarring (cirrhosis) when left unchecked. Liver stiffness measurements are used to diagnose fibrosis and scarring, and the median stiffness quadruples between normal and cirrhotic individuals (6.3 kPa versus 27.4 kPa) (*77*). Triggered by metabolic syndrome and obesity, dysbiosis in the gut microbiota and intestinal permeability are believed to enable leakage of microbial products and cell wall components into the circulation, which drive inflammation by activating hepatic stellate cells and KCs (*78*, *79*). In vivo studies show that the intestinal microbiota contribute to all three components of NASH: hepatic steatosis, inflammation, and fibrosis (*79*).

YAP and TAZ are both increased in abundance in NASH and are implicated in its pathogenesis with respect to inflammation in hepatocytes and macrophages (*8*, *80*). Hyperactive TAZ in the liver causes inflammation by promoting proinflammatory cytokine production and myeloid cell infiltration (*7*). In NASH, increased TAZ abundance in hepatocytes induces Indian hedgehog (*Ihh*)

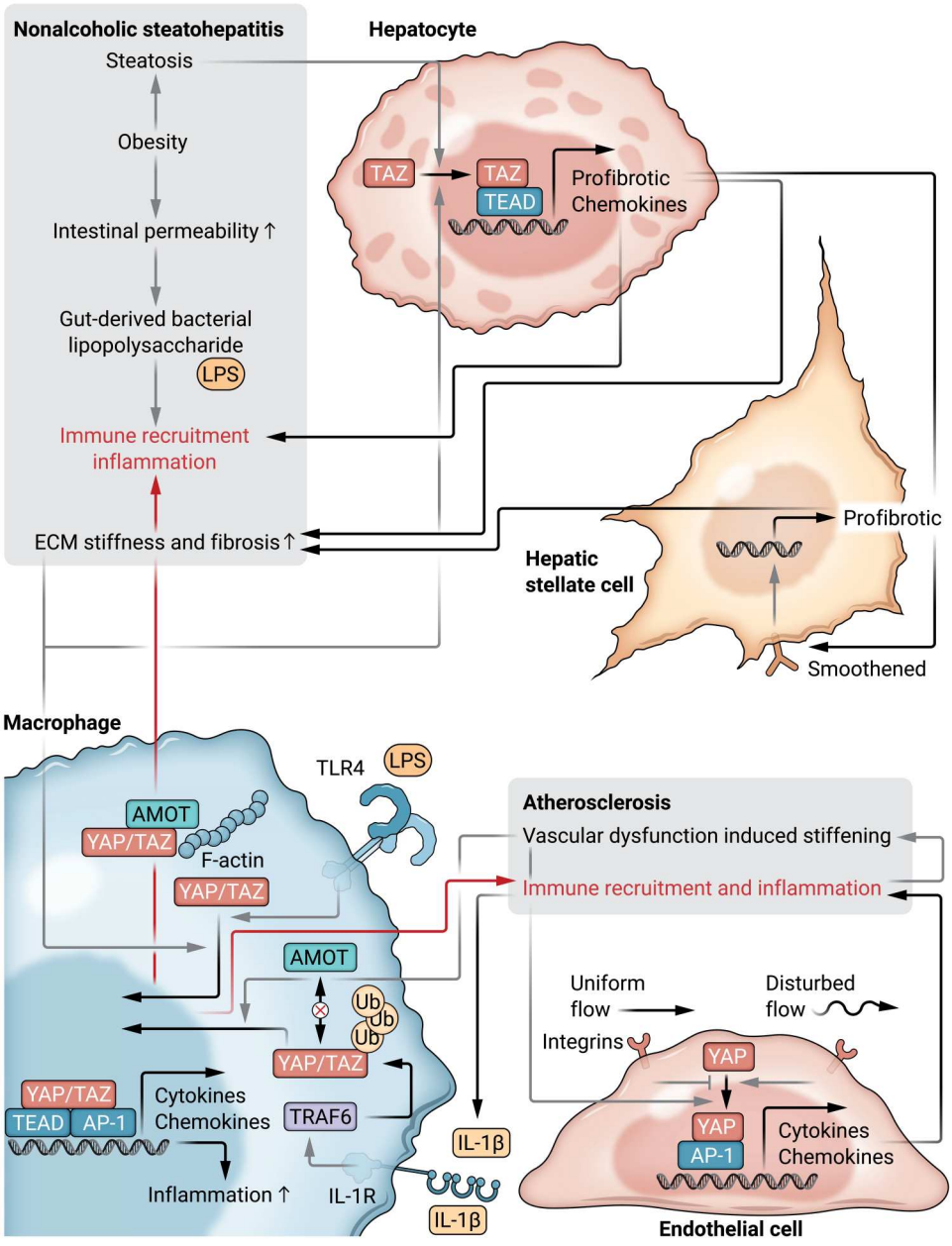

Fig. 3. Macrophage YAP/TAZ contribute to diseases by promoting chronic inflammation. YAP/TAZ activation in macrophages and other inflammation-capable cells, such as endothelial cells and hepatocytes, promotes inflammation in various diseases, including atherosclerosis and NASH. Local tissue stiffening and increased immune cell recruitment lead to a positive feedback loop that activates YAP/TAZ, which is, in turn, responsible for the production of proinflammatory cytokines, chemokines, and chemoattractants and the expression of profibrotic genes. Bolded typeface represents extracellular biophysical factors that modulate YAP/TAZ localization and cellular phenotype. Red typeface indicates pathogenic outcomes. Solid arrows indicate evidence for direct effects, whereas dashed arrows indicate evidence for indirect (cascading) effects.

expression, and Ihh subsequently activates the expression of fibrogenic genes in hepatic stellate cells (*8*). Hepatocyte-specific TAZ silencing through adeno-associated virus (AAV)–mediated shRNA delivery reverses inflammation, fibrosis, and hepatocellular death (*8*). Consistently, YAP knockout in KCs protects HFD-fed mice from hepatic inflammation and progression to NASH (*14*). In addition, constitutive YAP activation through 5SA-YAP expression in KCs enhances the production of proinflammatory cytokines, such as monocyte chemoattractant protein-1 (MCP-1), TNF-α, and IL-6. That study showed an increase in YAP enrichment at the promoters of genes encoding inflammatory cytokines upon inflammatory stimulation. It also proposed that LPS-induced inflammatory signaling promotes YAP nuclear translocation through histone deacetylase–mediated degradation of MST1. LPS-induced KC activation results in increased YAP abundance in a manner dependent on TLR4 and activator protein 1 (AP-1), implicating inflammatory

signaling in increasing YAP abundance. These results highlight the critical role of YAP in sustaining hepatic inflammation associated with obesity.

YAP deletion in macrophages does not suppress fibrosis or the expression of fibrogenic genes in NASH (*14*). It is possible that these genes are under the control of TAZ, as has been reported in hepatocytes (*8*). Note that macrophage-specific TAZ knockout results in reduced fibrosis and collagen deposition in models of renal fibrosis induced by unilateral ureter obstruction and ischemic/reperfusion injury (*54*). In addition, the influences of pathological fibrosis and consequent ECM stiffening on cellular mechanotransduction and YAP/TAZ activation in macrophages is yet to be examined. In this regard, it may be posited that YAP/TAZ activation in KCs can drive chronic inflammation by promoting fibrosis, and further studies are warranted to reveal the possible differential influences of YAP and TAZ in KCs.

Macrophage YAP/TAZ hamper tissue repair and regeneration

In several diseases, chronic inflammation impairs healing through delayed tissue regeneration. Alternatively activated (M2) macrophages drive tissue repair by producing pro-healing growth factors and cytokines. YAP/TAZ activity generally promotes tissue regeneration by inducing the expression of pro-proliferative genes in nonimmune cells. In contrast, YAP/TAZ activities in macrophages sustain disease and hamper healing through sustained inflammation. We highlight a few examples in which macrophage YAP/TAZ are responsible for curbing M2 macrophage activation and suppressing disease resolution (Fig. 4), whereas nonimmune cells may contribute to tissue regeneration or fibrosis because of the YAP/TAZ-dependent transcription of pro-proliferative or profibrotic genes. This highlights the complexity of YAP/TAZ roles in inflammatory diseases and the need for studies that dissect the contrasting contributions of YAP/TAZ in a cell-specific manner, as well as for therapeutics that target YAP/TAZ in specific subsets of cells.

Given the prominent roles of YAP/TAZ in keratinocyte proliferation, differentiation, and homeostasis (*81*), it is expected that dermal wound healing processes require YAP and TAZ. The topical application of YAP/TAZ-silencing RNA in a murine, full-thickness excision wound model results in delayed wound healing in the context of reduced transforming growth factor–β1 signaling, as exhibited by the reduced abundances of Smad2, p21, and Smad7 (*82*). YAP/TAZ silencing also causes reduced production of connective tissue growth factor (CTGF), contributing to the delayed healing (*82*). Importantly, YAP/TAZ are active (nuclear) in wounded dermis that is actively healing. In contrast, YAP/TAZ are both cytoplasmic and nuclear in uninjured tissue, suggesting that they exhibit a lower level of activity (*82*). Skin scar tissue can present with viscoelastic and nanomechanical properties that are very distinct from those of healthy intact tissue. For example, skin stiffness increases from a range of 1 to 20 kPa in healthy cases to ~50 kPa during the final stages of re-epithelialization and eventually to ~80 kPa during fibrosis (*83*). In addition to skin compliance, the dynamic mechanical forces associated with the different phases of dermal wound healing and ECM remodeling may also influence YAP/TAZ nucleocytoplasmic localization, contributing to cellular mechanosensing and adaptation, making it especially important to understand the effects of YAP/TAZ in macrophages participating in wound healing.

Macrophage function is a major determinant of wound healing outcomes and skin fibrosis. We found that in full-thickness wounds in mice that were treated with Tegaderm (a stiff dressing), the application of a fibrin gel led to a reduction in final scar size (*11*). Whereas there was no difference in macrophage infiltration between the fibrin- and Tegaderm-treated wounds, macrophages in contact with fibrin had substantially reduced amounts of inducible nitric oxide synthase (iNOS) and YAP (*11*). These results demonstrate the importance of modulating YAP activation in macrophages to reduce inflammation. In addition, a study described the role of YAP in wound fibroblasts during dermal scarring. YAP activity was involved in the activation of *En1*, a gene implicated in scarring and fibrosis (*84*). Fibroblast-specific silencing of YAP or treatment with verteporfin enhanced dermal regeneration by suppressing fibrosis and promoting secondary skin elements (*84*). Noting that this study did not focus on macrophages or inflammation, detailed investigations on the effects of macrophage-specific YAP/TAZ knockout on chronic wounds (such as diabetic wounds), dermal fibrosis, and infectious inflammatory wounds are warranted.

Inflammatory bowel disease (IBD) refers to a group of chronic inflammatory conditions of the gastrointestinal tract comprising Crohn's disease and ulcerative colitis. IBD represents a state of failed intestinal homeostasis illustrated by an increase in mucosal ECM breakdown and an altered microbiome that are associated with a slower rate of epithelial regeneration amidst sustained inflammation. In addition, IBD complications, such as intestinal fibrosis, are preceded by an increase in stiffness of between 55 and 65% (~900 to 1100 Pa in patients versus ~500 to 700 Pa in healthy controls), which also correlates with increased local inflammation (*85*). Curbing inflammation can assist in epithelial reconstitution, thereby alleviating the condition. Even under physiological conditions, intestinal tissue regeneration is vigorous. Regeneration-associated dynamic ECM remodeling generates extracellular biomechanical forces that activate YAP/TAZ through focal adhesion kinase– and Src-mediated actin polymerization (*86*, *87*). YAP/TAZ are required and sufficient to induce epithelial reprogramming in the context of dextran sulfate sodium (DSS)–induced colitis, where they act as mechanosensors that promote cellular proliferation (*86*). Inactivation of YAP/TAZ in the intestinal epithelium can prevent repair, as is evident by the reduced abundance of Sca-1, a marker of intestinal healing (*86*). Similar results were observed in another study that found that YAP was dispensable for normal intestinal homeostasis but critical for recovery after DSS-induced colitis (*88*). That study also showed that the dysregulation of the Hippo pathway by knockout of *Sav1* causes YAP hyperactivation and accelerated polyp formation in DSS-treated mice (*88*). This finding highlights a need for exquisite control over YAP/TAZ signaling to avoid unintended consequences physiologically.

As effector cells of the innate immune system, macrophages help maintain intestinal homeostasis. IBD resolution is driven by M2-polarized macrophages that suppress inflammation and support healing. A study showed that myeloid cell–specific knockout of YAP attenuates inflammation and promotes recovery after DSS-induced colitis (*13*). YAP abundance is influenced by the polarization state of macrophages. LPS induces increased YAP generation, whereas IL-4 and IL-13 induce a decrease in YAP abundance in macrophages. YAP also tunes the equilibrium between the M1 and M2 polarization of macrophages (*13*). The activation of YAP

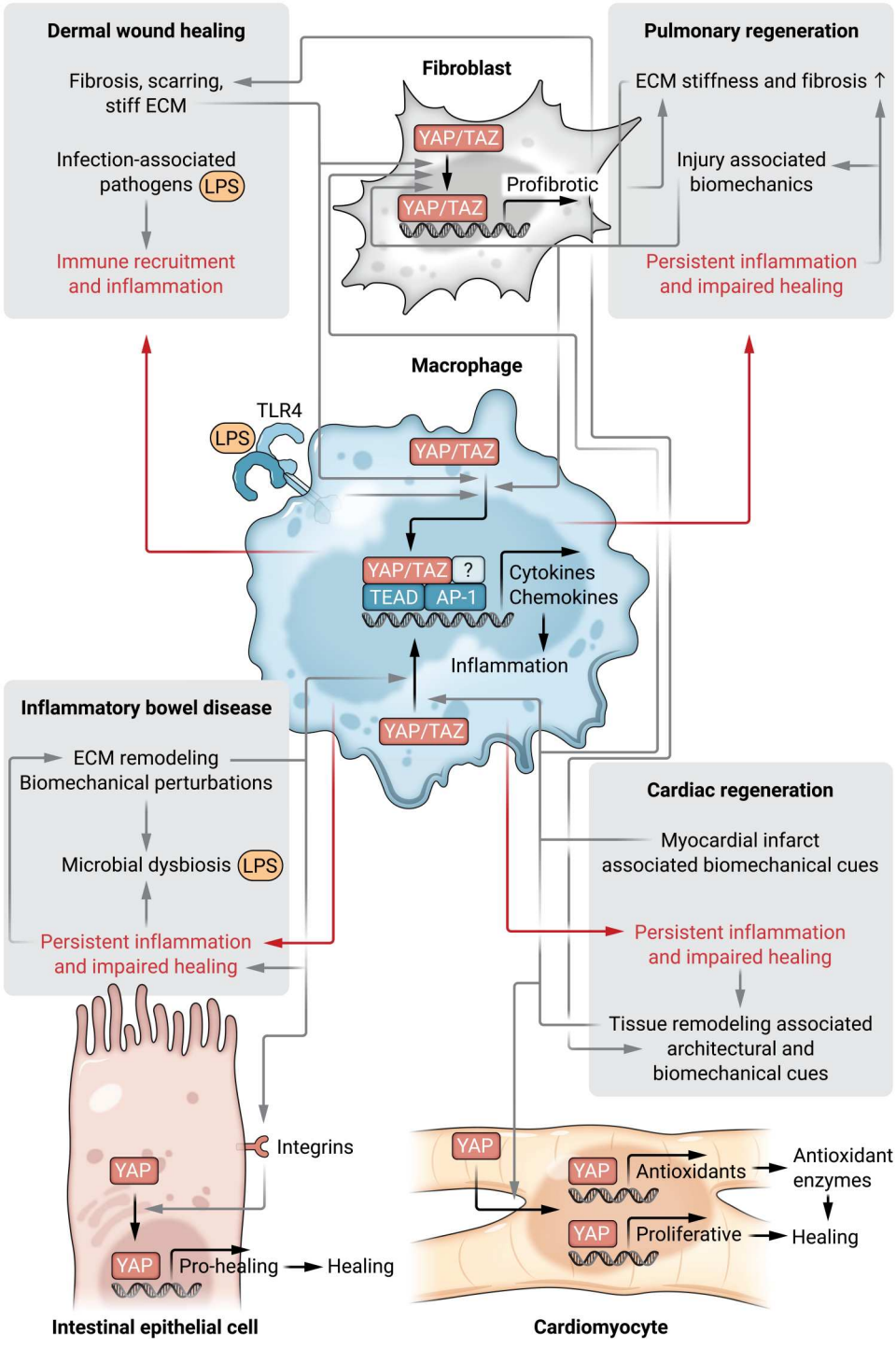

Fig. 4. Macrophage YAP/TAZ contribute to diseases through inflammation-associated impairment of regeneration. In conditions such as IBD and dermal, pulmonary, and cardiac regeneration, YAP/TAZ activation in tissue-resident, nonimmune cells resolves disease by promoting cellular proliferation. However, YAP/TAZ activation in fibroblasts and macrophages may have undesirable outcomes by promoting fibrosis and inflammation, respectively. Bolded typeface represents extracellular biophysical factors that modulate YAP/TAZ localization and cellular phenotype. Red typeface indicates pathogenic outcomes. Solid arrows indicated evidence for direct effects, whereas dashed arrows indicate evidence for indirect (cascading) effects.

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in macrophages in IBD stimulates IL-6 production and associated inflammation. Furthermore, YAP inhibits the activation of M2 macrophages by increasing the abundance of p53, causing a disequilibrium in the M1-M2 balance. Subsequently, activation of macrophage YAP triggers dysregulation of the gut microbiome. Myeloid cell–specific knockout of YAP enables DSS-treated mice to maintain an increased proportion of beneficial Bacteroidetes, *Lactobacillus*, and *Bifidobacteria*, in addition to suppressing potentially harmful microbes, such as Prevotellaceae (*13*). These results reveal the extent to which macrophage YAP enables the various pathological processes associated with IBD, making it an appealing target for therapy.

Cardiac development in the mouse fetus requires YAP because the loss of YAP results in lethal myocardial hypoplasia in the context of decreased cardiomyocyte proliferation (*89*). YAP knockdown also impedes regeneration and promotes fibrosis in the murine neonatal heart after myocardial infarction (MI) (*90*). YAP/TAZ are active in developing cardiac tissue, but this declines with age (*89*). As a result, murine postnatal (adult) hearts exhibit a limited ability for regeneration. Cardiac regeneration is accompanied by biomechanical forces arising from extensive ECM remodeling and architectural changes. In addition, fibrosis during post-MI regeneration results in scar tissue that is fourfold stiffer (55 kPa versus 18 kPa in healthy tissue) and exhibits impaired muscle distensibility and function (*91*). Accordingly, whether YAP activation can be therapeutically modulated to induce myocardial regeneration is being investigated.

The roles of YAP/TAZ in cardiac regeneration are cell type specific. Transgenic mice with a cardiac-specific YAP S112A mutation, which enhances Yap activity, exhibit enhanced cardiomyocyte proliferation, cardiac recovery, and survival after MI (*90*). A similar enhanced survival and recovery after MI is therapeutically induced by AAV-mediated, cardiac-specific YAP activation that stimulates cardiomyocyte proliferation, increases the expression of cell cycle genes, and promotes a less mature cardiac gene signature (*92*). YAP forms functional complexes with forkhead box protein O1 (FoxO1) in cardiomyocytes, enabling the transcription of genes encoding antioxidants (catalase and manganese superoxide dismutase) that protect cells from stress-induced death and injury from ischemia-reperfusion (*93*). Conversely, in macrophages and fibroblasts, YAP/TAZ activation stimulates inflammatory and fibrotic programs, respectively, thereby curbing cardiac regeneration after injuries, such as MI. After MI, cardiac fibroblasts experience increased YAP activation, which promotes fibrosis through the production of the profibrotic cytokine IL-33 (*94*). This is consistent with the observation that soft substrates prevent profibrotic fibroblast programs and enhance cardiac reprogramming by attenuating integrin, Rho-ROCK, actomyosin, and subsequent YAP/TAZ signaling (*95*). A fibroblast-specific loss of YAP/TAZ also causes reduced proinflammatory activation and chemokine-mediated recruitment of macrophages. YAP/TAZ activation in macrophages and the accompanying inflammation impair reparative responses (*12*). YAP activation causes IL-6 production while suppressing generation of the M2-marker Arg1, indicating a disequilibrium in the M1-M2 balance. Genetic deletion of YAP/TAZ enables reduced fibrosis and improved angiogenesis and survival in mice after MI-associated injury (*12*). These results highlight the cell type–specific effects of YAP/TAZ transcriptional programs, justifying the need for YAP/TAZ-targeted therapy to be aimed at specific cell populations.

Lung regeneration after pulmonary injury requires type I alveolar epithelial cells that line the alveoli to be replaced by progenitor type II alveolar epithelial cells. Pulmonary regeneration, such as after bleomycin-induced injury, requires TAZ activation for cellular differentiation and proliferation (*96*). The mechanical tension arising from regenerative processes activates YAP through F-actin and MAPK signaling, causing enhanced alveolar stem cell proliferation and restoration (*97*). The activation of YAP in lung fibroblasts induces the production of CTGF and type I collagen, resulting in a fibrotic response (*98*, *99*). Consistently, the fibroblastic foci of lungs from patients with idiopathic pulmonary fibrosis have an increased amount of TAZ (*99*). The increased expression of profibrotic YAP/TAZ targets in fibroblasts is believed to increase ECM stiffness and sustain YAP/TAZ activation in a feedback loop that underlies pulmonary fibrosis (*100*). Such fibrotic diseases can result in ~30-fold stiffer lungs compared with healthy tissue (20 to 100 kPa versus 1 to 5 kPa) (*101*). These results demonstrate a context-specific contribution of YAP/TAZ to pulmonary disease and recovery.

The prolonged use of mechanical ventilation in patients is associated with an immense risk of lung inflammation and injury. Recovery requires the activation of pro-healing, M2 macrophages. A study showed that YAP activation in alveolar macrophages causes production of the inflammatory cytokines TNF-α, IL-6, and IL-1β (*10*). Macrophage YAP deficiency attenuates inflammation and accelerates recovery by enhancing the polarization of M2 macrophages, whereas overexpression of YAP causes inflammation that inhibits regeneration (*10*). These results once again highlight the bias of YAP/TAZ transcriptional activity in macrophages toward M1 cells, resulting in sustained inflammation and delayed tissue restoration.

DISCUSSION

The role of YAP/TAZ in tissue growth and homeostasis has been long explored, but the ability of YAP/TAZ to influence immunity and inflammation has only recently become more perspicuous. Considering their transcriptional function to enhance inflammation by inducing the expression of target inflammatory genes and their nontranscriptional activity to control immunity, YAP/TAZ are good candidates for therapeutic intervention to combat various diseases and disorders. From various studies, it is clear that YAP and TAZ are spatiotemporally distinct, have tissue-specific roles, and function in a context-dependent manner. Therefore, targeting YAP and TAZ similarly in every tissue may not be the best therapeutic approach. Some of the possible approaches to target YAP/TAZ in cells include, but are not limited to, pharmacological regulation, genetic manipulation, and biomaterial-based interventions.

Pharmacological regulation of YAP/TAZ aims to curb aberrant biomechanically activated transcriptional programs. The positive feedback loop involving pathological tissue stiffening, increased intracellular tension, YAP/TAZ nuclear translocation, and consequential expression of profibrotic genes leading to fibrosis is one such example of the dynamic reciprocity between cells and the ECM that underlies several diseases. Verteporfin is an U.S. Food and Drug Administration (FDA)–approved drug that is used as a photosensitizer in photodynamic therapy for choroidal blood

vessel ablation in age-related macular degeneration. Its function as a YAP/TAZ inhibitor is light independent, whereby it targets YAP/TAZ-TEAD complex formation to inhibit target gene expression (*102*). The effectiveness of verteporfin has been demonstrated in several fibrotic diseases, including those of the liver, lung, and skin (*84*, *103*, *104*). Dimethyl fumarate (DMF) is effective in treating systemic sclerosis, a fibrotic disease characterized by vascular, immune, and fibrotic changes in multiple organs (*105*). DMF reduces the nuclear localization of YAP/TAZ and causes antifibrotic effects by inhibiting the phosphatidylinositol 3-kinase–Akt1 pathway. FDA-approved statins are also promising because they target YAP/TAZ nuclear localization through inhibition of the Rho and mevalonate pathways (*106*, *107*). In addition to these drugs, a peptide mimic of vestigial-like family member 4 (VGLL4) inhibits YAP/TAZ-mediated transcriptional activity in cancer by competitive binding to TEADs (*108*). Such peptide mimics of the binding domains of VGLL4, AT-rich interaction domain 1A (ARID1A) (*109*), and YAP can disrupt YAP/TAZ-TEAD interactions and have the potential to treat inflammatory diseases (*110*). However, systemic inhibition of YAP/TAZ may have detrimental effects in many pathologies because of the roles of YAP/TAZ in healing-associated cellular proliferation. Genetic therapies may instead be used to control YAP/TAZ abundance and activation in a more targeted, cell type–specific, and context-dependent manner.

AAV-based *YAP1* gene therapy improves cardiac recovery after MI in mice (*92*). The study used a cardiac-specific, inducible system to express and activate YAP in cardiomyocytes, resulting in cellular proliferation and accelerated recovery as exhibited by improved cardiac function and survival. AAV-mediated therapeutics can target specific tissues because of capsid serotype–driven tropism, and therapeutic genes can be expressed in specific cell types by placing them under the control of cell type–specific regulatory elements (*111*). Such targeted and inducible systems can be used to selectively switch on YAP/TAZ activity at those phases of recovery that require the proliferation of specific subsets of cells. In one study, mechanoresponsive cellular systems were engineered that exploit YAP/TAZ as biosensors to activate drugs selectively. Cytosine deaminase was exogenously induced by a YAP/TAZ-responsive element in mesenchymal stem cells, which enabled these cells to produce the enzyme when they sensed stiffer microenvironments, which led to conversion of the prodrug 5-fluorocytosine to the active drug form 5-fluorouracil (*112*). Although such systems were developed for cancer treatment (*113*), we posit that similar systems may be designed to fastidiously target stiffer microenvironments with anti-inflammatory or pro-healing therapeutics using YAP/TAZ as mechanosensors.

Biomaterial design can also benefit from leveraging the modulation of immune cells by YAP/TAZ. Biomaterial implantation induces inflammatory and fibrotic reactions through the foreign body reaction. During this process, immune cells, including macrophages, are recruited to the implant site. Because macrophages are among the first inflammatory immune cells to be recruited, it is beneficial to suppress their inflammatory activation by modifying the properties of the biomaterials and implants. Similarly, we found that treating wounds with softer fibrin hydrogels reduced scar size and suppressed YAP production in macrophages in a full-thickness wound model (*11*). Furthermore, we demonstrated that in the tissue surrounding a stiff hydrogel implant, macrophages had increased amounts of iNOS and YAP. These findings suggest that it might be possible to control the nucleo-cytoplasmic shuttling of YAP/TAZ by manipulating biomaterial properties, thereby controlling inflammation. Another promising strategy to modulate macrophage inflammatory activation is changing their cell shape and, possibly, those of other immune cells by altering the surface properties of the biomaterial. By modifying the surface topography of the material (biomimetic multiscale wrinkles and micro- and nanopatterned groves) (*114*, *115*), porosity (*116*), or micropatterning technique (*117*), it is possible to change the cell shape and polarize the macrophage toward an alternatively activated phenotype. These shape changes associated with reduced inflammation occur through modulation of Src, a kinase that is upstream of YAP/TAZ (*45*). More studies are warranted to pinpoint the role of cell shape in regulating YAP/TAZ in innate immune cells. Overall, YAP/TAZ can be strong determinants of the inflammatory behavior of innate immune cells that can be controlled by rational biomaterial design.

Whereas several studies showed increased M1 polarization, inflammation, intracellular calcium signaling, and phagocytosis in macrophages on stiffer substrates (*11*, *61*, *118*–*123*), findings from other studies contradict the proinflammatory effects of stiffer substrates by observing increased inflammasome activation and IL-1β production in cells on compliant surfaces (*124*–*126*). Some of these differences may be due to different in vitro experimental conditions (such as cell types and origin, ECM types, substrate chemistry/architecture, two-dimensional versus three-dimensional culture, cell density, and time of adhesion). Interestingly, cytoplasmic YAP/TAZ are implicated in inflammasome stabilization (*60*). YAP/TAZ largely remain cytoplasmic in cells adhered to softer surfaces, explaining the observed greater inflammasome activation in cells on compliant substrates. YAP/TAZ can play multifaceted roles depending on their location, having proinflammatory transcriptional roles in the nucleus but mediating inflammasome activation/stabilization (*60*) and suppression of the antiviral response (*63*–*65*) in the cytoplasm. These findings warrant a holistic study of the effects of YAP/TAZ on macrophage-mediated inflammation and disease progression, with a focus on the effects of differential YAP/TAZ activation, temporal localization (shuttling), and abundance.

Last, one of the most important challenges in targeting YAP/TAZ therapeutically is their cell-specific expression and context-dependent function. As we described earlier, suppressing YAP/TAZ can have either beneficial or undesirable pathophysiological outcomes depending on the cell type being targeted. Briefly, YAP/TAZ have inflammatory roles not only in immune cells but also in cell types such as endothelial cells and hepatocytes, which propagate inflammatory programs. In structural cells, such as epithelial cells and fibroblasts, YAP/TAZ activation either promotes healing through cellular proliferation or is responsible for pathological conditions, such as fibrosis. These findings underline the necessity to manipulate YAP/TAZ in a cell type–specific and temporal manner for desired therapeutic outcomes in mechano-inflammatory pathologies. Cell and gene therapies could help customize YAP/TAZ-targeted therapeutics to affect only the cell populations of interest in a disease-specific manner. Further understanding of YAP/TAZ roles in immune cells that are mechanosensory would assist in tailoring such therapeutic regimens and biomaterials to promote better health outcomes.

REFERENCES AND NOTES

1. T. Panciera, L. Azzolin, M. Cordenonsi, S. Piccolo, Mechanobiology of YAP and TAZ in physiology and disease. *Nat. Rev. Mol. Cell Biol.* **18**, 758–770 (2017).
2. F. X. Yu, B. Zhao, K. L. Guan, Hippo pathway in organ size control, tissue homeostasis, and cancer. *Cell* **163**, 811–828 (2015).
3. N. Wang, Review of cellular mechanotransduction. *J. Phys. D Appl. Phys.* **50**, 233002 (2017).
4. G. Brusatin, T. Panciera, A. Gandin, A. Citron, S. Piccolo, Biomaterials and engineered microenvironments to control YAP/TAZ-dependent cell behaviour. *Nat. Mater.* **17**, 1063–1075 (2018).
5. A. Pocaterra, P. Romani, S. Dupont, YAP/TAZ functions and their regulation at a glance. *J. Cell Sci.* **133**, jcs230425 (2020).
6. J. Francisco, Y. Zhang, Y. Nakada, J. I. Jeong, C. Y. Huang, A. Ivessa, S. Oka, G. J. Babu, D. P. Del Re, AAV-mediated YAP expression in cardiac fibroblasts promotes inflammation and increases fibrosis. *Sci. Rep.* **11**, 10553 (2021).
7. T. J. Hagenbeek, J. D. Webster, N. M. Kljavin, M. T. Chang, T. Pham, H. J. Lee, C. Klijn, A. G. Cai, K. Totpal, B. Ravishankar, N. Yang, D. H. Lee, K. B. Walsh, G. Hatzivassiliou, C. C. de la Cruz, S. E. Gould, X. Wu, W. P. Lee, S. Yang, Z. Zhang, Q. Gu, Q. Ji, E. L. Jackson, D. S. Lim, A. Dey, The Hippo pathway effector TAZ induces TEAD-dependent liver inflammation and tumors. *Sci. Signal.* **11**, eaaaj1757 (2018).
8. X. Wang, Z. Zheng, J. M. Caviglia, K. E. Corey, T. M. Herfel, B. Cai, R. Masia, R. T. Chung, J. H. Lefkowitch, R. F. Schwabe, I. Tabas, Hepatocyte TAZ/WWTR1 promotes inflammation and fibrosis in nonalcoholic steatohepatitis. *Cell Metab.* **24**, 848–862 (2016).
9. M. Liu, M. Yan, H. Lv, B. Wang, X. Lv, H. Zhang, S. Xiang, J. Du, T. Liu, Y. Tian, X. Zhang, F. Zhou, T. Cheng, Y. Zhu, H. Jiang, Y. Cao, D. Ai, Macrophage K63-linked ubiquitination of YAP promotes its nuclear localization and exacerbates atherosclerosis. *Cell Rep.* **32**, 107990 (2020).
10. Q. Luo, J. Luo, Y. Wang, YAP deficiency attenuates pulmonary injury following mechanical ventilation through the regulation of M1/M2 macrophage polarization. *J. Inflamm. Res.* **13**, 1279–1290 (2020).
11. V. S. Meli, H. Atcha, P. K. Veerasubramanian, R. R. Nagalla, T. U. Luu, E. Y. Chen, C. F. Guerrero-Juarez, K. Yamaga, W. Pandori, J. Y. Hsieh, T. L. Downing, D. A. Fruman, M. B. Lodoen, M. V. Plikus, W. Wang, W. F. Liu, YAP-mediated mechanotransduction tunes the macrophage inflammatory response. *Sci. Adv.* **6**, eabb8471 (2020).
12. M. M. Mia, D. M. Cibi, S. A. B. Abdul Ghani, W. Song, N. Tee, S. Ghosh, J. Mao, E. N. Olson, M. K. Singh, YAP/TAZ deficiency reprograms macrophage phenotype and improves infarct healing and cardiac function after myocardial infarction. *PLOS Biol.* **18**, e3000941 (2020).
13. X. Zhou, W. Li, S. Wang, P. Zhang, Q. Wang, J. Xiao, C. Zhang, X. Zheng, X. Xu, S. Xue, L. Hui, H. Ji, B. Wei, H. Wang, YAP aggravates inflammatory bowel disease by regulating M1/M2 macrophage polarization and gut microbial homeostasis. *Cell Rep.* **27**, 1176–1189.e5 (2019).
14. K. Song, H. Kwon, C. Han, W. Chen, J. Zhang, W. Ma, S. Dash, C. R. Gandhi, T. Wu, Yes-associated protein in Kupffer cells enhances the production of proinflammatory cytokines and promotes the development of nonalcoholic steatohepatitis. *Hepatology* **72**, 72–87 (2020).
15. S. Wang, L. Zhou, L. Ling, X. Meng, F. Chu, S. Zhang, F. Zhou, The crosstalk between hippo-YAP pathway and innate immunity. *Front. Immunol.* **11**, 323 (2020).
16. T. Moroishi, C. G. Hansen, K. L. Guan, The emerging roles of YAP and TAZ in cancer. *Nat. Rev. Cancer* **15**, 73–79 (2015).
17. F. X. Yu, K. L. Guan, The Hippo pathway: Regulators and regulations. *Genes Dev.* **27**, 355–371 (2013).
18. L. Chen, Non-canonical Hippo signaling regulates immune responses. *Adv. Immunol.* **144**, 87–119 (2019).
19. L. An, P. Nie, M. Chen, Y. Tang, H. Zhang, J. Guan, Z. Cao, C. Hou, W. Wang, Y. Zhao, H. Xu, S. Jiao, Z. Zhou, MST4 kinase suppresses gastric tumorigenesis by limiting YAP activation via a non-canonical pathway. *J. Exp. Med.* **217**, e20191817 (2020).
20. Y. S. Cho, J. Jiang, Hippo-independent regulation of Yki/Yap/Taz: A non-canonical view. *Front. Cell Dev. Biol.* **9**, 658481 (2021).
21. M. Huse, Mechanical forces in the immune system. *Nat. Rev. Immunol.* **17**, 679–690 (2017).
22. M. Sudol, P. Bork, A. Einbond, K. Kastury, T. Druck, M. Negrini, K. Huebner, D. Lehman, Characterization of the mammalian YAP (Yes-associated protein) gene and its role in defining a novel protein module, the WW domain. *J. Biol. Chem.* **270**, 14733–14741 (1995).
23. J. E. Thaventhiran, A. Hoffmann, L. Magiera, M. de la Roche, H. Lingel, M. Brunner-Weinzierl, D. T. Fearon, Activation of the Hippo pathway by CTLA-4 regulates the expression of Blimp-1 in the CD8+ T cell. *Proc. Natl. Acad. Sci. U.S.A.* **109**, E2223–E2229 (2012).
24. Q. Zhang, F. Meng, S. Chen, S. W. Plouffe, S. Wu, S. Liu, X. Li, R. Zhou, J. Wang, B. Zhao, J. Liu, J. Qin, J. Zou, X. H. Feng, K. L. Guan, P. Xu, Hippo signalling governs cytosolic nucleic acid sensing through YAP/TAZ-mediated TBK1 blockade. *Nat. Cell Biol.* **19**, 362–374 (2017).
25. M. Karlsson, C. Zhang, L. Mear, W. Zhong, A. Digre, B. Katona, E. Sjostedt, L. Butler, J. Odeberg, P. Dusart, F. Edfors, P. Oksvold, K. von Feilitzen, M. Zwahlen, M. Arif, O. Altay, X. Li, M. Ozcan, A. Mardinoglu, L. Fagerberg, J. Mulder, Y. Luo, F. Ponten, M. Uhlen, C. Lindskog, A single-cell type transcriptomics map of human tissues. *Sci. Adv.* **7**, eabh2169 (2021).
26. Z. Meng, Y. Qiu, K. C. Lin, A. Kumar, J. K. Placone, C. Fang, K. C. Wang, S. Lu, M. Pan, A. W. Hong, T. Moroishi, M. Luo, S. W. Plouffe, Y. Diao, Z. Ye, H. W. Park, X. Wang, F. X. Yu, S. Chien, C. Y. Wang, B. Ren, A. J. Engler, K. L. Guan, RAP2 mediates mechanoresponses of the Hippo pathway. *Nature* **560**, 655–660 (2018).
27. I. Dasgupta, D. McCollum, Control of cellular responses to mechanical cues through YAP/TAZ regulation. *J. Biol. Chem.* **294**, 17693–17706 (2019).
28. A. Totaro, T. Panciera, S. Piccolo, YAP/TAZ upstream signals and downstream responses. *Nat. Cell Biol.* **20**, 888–899 (2018).
29. K. Wada, K. Itoga, T. Okano, S. Yonemura, H. Sasaki, Hippo pathway regulation by cell morphology and stress fibers. *Development* **138**, 3907–3914 (2011).
30. M. Kim, M. Kim, S. Lee, S. Kuninaka, H. Saya, H. Lee, S. Lee, D. S. Lim, cAMP/PKA signalling reinforces the LATS-YAP pathway to fully suppress YAP in response to actin cytoskeletal changes. *EMBO J.* **32**, 1543–1555 (2013).
31. M. Aragona, T. Panciera, A. Manfrin, S. Giulitti, F. Michielin, N. Elvassore, S. Dupont, S. Piccolo, A mechanical checkpoint controls multicellular growth through YAP/TAZ regulation by actin-processing factors. *Cell* **154**, 1047–1059 (2013).
32. A. Das, R. S. Fischer, D. Pan, C. M. Waterman, YAP nuclear localization in the absence of cell-cell contact is mediated by a filamentous actin-dependent, myosin II- and phospho-YAP-independent pathway during extracellular matrix mechanosensing. *J. Biol. Chem.* **291**, 6096–6110 (2016).
33. S. Dupont, L. Morsut, M. Aragona, E. Enzo, S. Giulitti, M. Cordenonsi, F. Zanconato, J. Le Digabel, M. Forcato, S. Bicciato, N. Elvassore, S. Piccolo, Role of YAP/TAZ in mechanotransduction. *Nature* **474**, 179–183 (2011).
34. Q. Li, S. Li, S. Mana-Capelli, R. J. Roth Flach, L. V. Danai, A. Amcheslavsky, Y. Nie, S. Kaneko, X. Yao, X. Chen, J. L. Cotton, J. Mao, D. McCollum, J. Jiang, M. P. Czech, L. Xu, Y. T. Ip, The conserved misshapen-warts-Yorkie pathway acts in enteroblasts to regulate intestinal stem cells in Drosophila. *Dev. Cell* **31**, 291–304 (2014).
35. Z. Meng, T. Moroishi, V. Mottier-Pavie, S. W. Plouffe, C. G. Hansen, A. W. Hong, H. W. Park, J. S. Mo, W. Lu, S. Lu, F. Flores, F. X. Yu, G. Halder, K. L. Guan, MAP4K family kinases act in parallel to MST1/2 to activate LATS1/2 in the Hippo pathway. *Nat. Commun.* **6**, 8357 (2015).
36. Y. Zheng, W. Wang, B. Liu, H. Deng, E. Uster, D. Pan, Identification of happyhour/MAP4K as alternative Hpo/Mst-like kinases in the hippo kinase cascade. *Dev. Cell* **34**, 642–655 (2015).
37. S. W. Chan, C. J. Lim, F. Guo, I. Tan, T. Leung, W. Hong, Actin-binding and cell proliferation activities of angiomotin family members are regulated by Hippo pathway-mediated phosphorylation. *J. Biol. Chem.* **288**, 37296–37307 (2013).
38. S. Mana-Capelli, D. McCollum, Angiomotins stimulate LATS kinase autophosphorylation and act as scaffolds that promote Hippo signaling. *J. Biol. Chem.* **293**, 18230–18241 (2018).
39. S. Mana-Capelli, M. Paramasivam, S. Dutta, D. McCollum, Angiomotins link F-actin architecture to Hippo pathway signaling. *Mol. Biol. Cell* **25**, 1676–1685 (2014).
40. Y. Li, H. Zhou, F. Li, S. W. Chan, Z. Lin, Z. Wei, Z. Yang, F. Guo, C. J. Lim, W. Xing, Y. Shen, W. Hong, J. Long, M. Zhang, Angiomotin binding-induced activation of Merlin/NF2 in the Hippo pathway. *Cell Res.* **25**, 801–817 (2015).
41. P. Li, M. R. Silvis, Y. Honaker, W. H. Lien, S. T. Arron, V. Vasioukhin, αE-catenin inhibits a Src-YAP1 oncogenic module that couples tyrosine kinases and the effector of Hippo signaling pathway. *Genes Dev.* **30**, 798–811 (2016).
42. J. Rosenbluh, D. Nijhawan, A. G. Cox, X. Li, J. T. Neal, E. J. Schafer, T. I. Zack, X. Wang, A. Tsherniak, A. C. Schinzel, D. D. Shao, S. E. Schumacher, B. A. Weir, F. Vazquez, G. S. Cowley, D. E. Root, J. P. Mesirov, R. Beroukhim, C. J. Kuo, W. Goessling, W. C. Hahn, β-catenin-driven cancers require a YAP1 transcriptional complex for survival and tumorigenesis. *Cell* **151**, 1457–1473 (2012).
43. Y. Si, X. Ji, X. Cao, X. Dai, L. Xu, H. Zhao, X. Guo, H. Yan, H. Zhang, C. Zhu, Q. Zhou, M. Tang, Z. Xia, L. Li, Y. S. Cong, S. Ye, T. Liang, X. H. Feng, B. Zhao, Src inhibits the hippo tumor suppressor pathway through tyrosine phosphorylation of Lats1. *Cancer Res.* **77**, 4868–4880 (2017).
44. K. Taniguchi, L. W. Wu, S. I. Grivennikov, P. R. de Jong, I. Lian, F. X. Yu, K. Wang, S. B. Ho, B. S. Boland, J. T. Chang, W. J. Sandborn, G. Hardiman, E. Raz, Y. Maehara, A. Yoshimura, J. Zucman-Rossi, K. L. Guan, M. Karin, A gp130-Src-YAP module links inflammation to epithelial regeneration. *Nature* **519**, 57–62 (2015).

45. P. K. Veerasubramanian, H. Shao, V. S. Meli, T. A. Q. Phan, T. U. Luu, W. F. Liu, T. L. Downing, A Src-H3 acetylation signaling axis integrates macrophage mechanosensation with inflammatory response. *Biomaterials* **279**, 121236 (2021).
46. K. T. Furukawa, K. Yamashita, N. Sakurai, S. Ohno, The epithelial circumferential actin belt regulates YAP/TAZ through nucleocytoplasmic shuttling of merlin. *Cell Rep.* **20**, 1435–1447 (2017).
47. F. D. Camargo, S. Gokhale, J. B. Johnnidis, D. Fu, G. W. Bell, R. Jaenisch, T. R. Brummelkamp, YAP1 increases organ size and expands undifferentiated progenitor cells. *Curr. Biol.* **17**, 2054–2060 (2007).
48. X. Cao, S. L. Pfaff, F. H. Gage, YAP regulates neural progenitor cell number via the TEA domain transcription factor. *Genes Dev.* **22**, 3320–3334 (2008).
49. I. Lian, J. Kim, H. Okazawa, J. Zhao, B. Zhao, J. Yu, A. Chinnaiyan, M. A. Israel, L. S. Goldstein, R. Abujarour, S. Ding, K. L. Guan, The role of YAP transcription coactivator in regulating stem cell self-renewal and differentiation. *Genes Dev.* **24**, 1106–1118 (2010).
50. L. Jansson, J. Larsson, Normal hematopoietic stem cell function in mice with enforced expression of the Hippo signaling effector YAP1. *PLOS ONE* **7**, e32013 (2012).
51. L. Zhao, H. Guan, C. Song, Y. Wang, C. Liu, C. Cai, H. Zhu, H. Liu, L. Zhao, J. Xiao, YAP1 is essential for osteoclastogenesis through a TEADs-dependent mechanism. *Bone* **110**, 177–186 (2018).
52. W. J. Boyle, W. S. Simonet, D. L. Lacey, Osteoclast differentiation and activation. *Nature* **423**, 337–342 (2003).
53. X. Du, J. Wen, Y. Wang, P. W. F. Karmaus, A. Khatamian, H. Tan, Y. Li, C. Guy, T. M. Nguyen, Y. Dhungana, G. Neale, J. Peng, J. Yu, H. Chi, Hippo/Mst signalling couples metabolic state and immune function of CD8α^+ dendritic cells. *Nature* **558**, 141–145 (2018).
54. Y. Feng, Y. Liang, X. Zhu, M. Wang, Y. Gui, Q. Lu, M. Gu, X. Xue, X. Sun, W. He, J. Yang, R. L. Johnson, C. Dai, The signaling protein Wnt5a promotes TGFβ1-mediated macrophage polarization and kidney fibrosis by inducing the transcriptional regulators Yap/Taz. *J. Biol. Chem.* **293**, 19290–19302 (2018).
55. K. Newton, V. M. Dixit, Signaling in innate immunity and inflammation. *Cold Spring Harb. Perspect. Biol.* **4**, a0006049 (2012).
56. L. Chen, H. Deng, H. Cui, J. Fang, Z. Zuo, J. Deng, Y. Li, X. Wang, L. Zhao, Inflammatory responses and inflammation-associated diseases in organs. *Oncotarget* **9**, 7204–7218 (2018).
57. T. Yamauchi, T. Moroishi, Hippo pathway in mammalian adaptive immune system. *Cell* **8**, 398 (2019).
58. L. Hong, X. Li, D. Zhou, J. Geng, L. Chen, Role of Hippo signaling in regulating immunity. *Cell. Mol. Immunol.* **15**, 1003–1009 (2018).
59. B. Liu, Y. Zheng, F. Yin, J. Yu, N. Silverman, D. Pan, Toll receptor-mediated hippo signaling controls innate immunity in Drosophila. *Cell* **164**, 406–419 (2016).
60. D. Wang, Y. Zhang, X. Xu, J. Wu, Y. Peng, J. Li, R. Luo, L. Huang, L. Liu, S. Yu, N. Zhang, B. Lu, K. Zhao, YAP promotes the activation of NLRP3 inflammasome via blocking K27-linked polyubiquitination of NLRP3. *Nat. Commun.* **12**, 2674 (2021).
61. M. L. Previtera, A. Sengupta, Substrate stiffness regulates proinflammatory mediator production through TLR4 activity in macrophages. *PLOS ONE* **10**, e0145813 (2015).
62. M. Chakraborty, K. Chu, A. Shrestha, X. S. Revelo, X. Zhang, M. J. Gold, S. Khan, M. Lee, C. Huang, M. Akbari, F. Barrow, Y. T. Chan, H. Lei, N. K. Kotoulas, J. Jovel, C. Pastrello, M. Kotlyar, C. Goh, E. Michelakis, X. Clemente-Casares, P. S. Ohashi, E. G. Engleman, S. Winer, I. Jurisica, S. Tsai, D. A. Winer, Mechanical stiffness controls dendritic cell metabolism and function. *Cell Rep.* **34**, 108609 (2021).
63. S. Wang, F. Xie, F. Chu, Z. Zhang, B. Yang, T. Dai, L. Gao, L. Wang, L. Ling, J. Jia, H. van Dam, J. Jin, L. Zhang, F. Zhou, YAP antagonizes innate antiviral immunity and is targeted for lysosomal degradation through IKKε-mediated phosphorylation. *Nat. Immunol.* **18**, 733–743 (2017).
64. F. Meng, R. Zhou, S. Wu, Q. Zhang, Q. Jin, Y. Zhou, S. W. Plouffe, S. Liu, H. Song, Z. Xia, B. Zhao, S. Ye, X. H. Feng, K. L. Guan, J. Zou, P. Xu, Mst1 shuts off cytosolic antiviral defense through IRF3 phosphorylation. *Genes Dev.* **30**, 1086–1100 (2016).
65. A. Aravamudhan, A. J. Haak, K. M. Choi, J. A. Meridew, N. Caporarello, D. L. Jones, Q. Tan, G. Ligresti, D. J. Tschumperlin, TBK1 regulates YAP/TAZ and fibrogenic fibroblast activation. *Am. J. Physiol. Lung Cell. Mol. Physiol.* **318**, L852–L863 (2020).
66. F. Zanconato, M. Forcato, G. Battilana, L. Azzolin, E. Quaranta, B. Bodega, A. Rosato, S. Bicciato, M. Cordenonsi, S. Piccolo, Genome-wide association between YAP/TAZ/TEAD and AP-1 at enhancers drives oncogenic growth. *Nat. Cell Biol.* **17**, 1218–1227 (2015).
67. C. Stein, A. F. Bardet, G. Roma, S. Bergling, I. Clay, A. Ruchti, C. Agarinis, T. Schmelzle, T. Bouwmeester, D. Schubeler, A. Bauer, YAP1 exerts its transcriptional control via TEAD-mediated activation of enhancers. *PLOS Genet.* **11**, e1005465 (2015).
68. F. Zanconato, G. Battilana, M. Forcato, L. Filippi, L. Azzolin, A. Manfrin, E. Quaranta, D. Di Biagio, G. Sigismondo, V. Guzzardo, P. Lejeune, B. Haendler, J. Krijgsveld, M. Fassan, S. Bicciato, M. Cordenonsi, S. Piccolo, Transcriptional addiction in cancer cells is mediated by YAP/TAZ through BRD4. *Nat. Med.* **24**, 1599–1610 (2018).
69. Y. Wang, K. Tu, D. Liu, L. Guo, Y. Chen, Q. Li, J. L. Maiers, Z. Liu, V. H. Shah, C. Dou, D. Tschumperlin, L. Voneschen, R. Yang, N. Kang, p300 acetyltransferase is a cytoplasm-to-nucleus shuttle for SMAD2/3 and TAZ nuclear transport in transforming growth factor β–stimulated hepatic stellate cells. *Hepatology* **70**, 1409–1423 (2019).
70. M. E. Gerritsen, A. J. Williams, A. S. Neish, S. Moore, Y. Shi, T. Collins, CREB-binding protein/p300 are transcriptional coactivators of p65. *Proc. Natl. Acad. Sci. U.S.A.* **94**, 2927–2932 (1997).
71. E. Nicodeme, K. L. Jeffrey, U. Schaefer, S. Beinke, S. Dewell, C. W. Chung, R. Chandwani, I. Marazzi, P. Wilson, H. Coste, J. White, J. Kirilovsky, C. M. Rice, J. M. Lora, R. K. Prinjha, K. Lee, A. Tarakhovsky, Suppression of inflammation by a synthetic histone mimic. *Nature* **468**, 1119–1123 (2010).
72. L. Wang, J. Y. Luo, B. Li, X. Y. Tian, L. J. Chen, Y. Huang, J. Liu, D. Deng, C. W. Lau, S. Wan, D. Ai, K. K. Mak, K. K. Tong, K. M. Kwan, N. Wang, J. J. Chiu, Y. Zhu, Y. Huang, Integrin-YAP/TAZ-JNK cascade mediates atheroprotective effect of unidirectional shear flow. *Nature* **540**, 579–582 (2016).
73. K. S. Cunningham, A. I. Gotlieb, The role of shear stress in the pathogenesis of atherosclerosis. *Lab. Invest.* **85**, 9–23 (2005).
74. A. Gotschy, E. Bauer, C. Schrodt, G. Lykowsky, Y. X. Ye, E. Rommel, P. M. Jakob, W. R. Bauer, V. Herold, Local arterial stiffening assessed by MRI precedes atherosclerotic plaque formation. *Circ. Cardiovasc. Imaging* **6**, 916–923 (2013).
75. K. C. Wang, Y. T. Yeh, P. Nguyen, E. Limqueco, J. Lopez, S. Thorossian, K. L. Guan, Y. J. Li, S. Chien, Flow-dependent YAP/TAZ activities regulate endothelial phenotypes and atherosclerosis. *Proc. Natl. Acad. Sci. U.S.A.* **113**, 11525–11530 (2016).
76. P. Yuan, Q. Hu, X. He, Y. Long, X. Song, F. Wu, Y. He, X. Zhou, Laminar flow inhibits the Hippo/YAP pathway via autophagy and SIRT1-mediated deacetylation against atherosclerosis. *Cell Death Dis.* **11**, 141 (2020).
77. N. Ganne-Carrie, M. Ziol, V. de Ledinghen, C. Douvin, P. Marcellin, L. Castera, D. Dhumeaux, J. C. Trinchet, M. Beaugrand, Accuracy of liver stiffness measurement for the diagnosis of cirrhosis in patients with chronic liver diseases. *Hepatology* **44**, 1511–1517 (2006).
78. Z. Safari, P. Gerard, The links between the gut microbiome and non-alcoholic fatty liver disease (NAFLD). *Cell. Mol. Life Sci.* **76**, 1541–1558 (2019).
79. M. Mouzaki, E. M. Comelli, B. M. Arendt, J. Bonengel, S. K. Fung, S. E. Fischer, I. D. McGilvray, J. P. Allard, Intestinal microbiota in patients with nonalcoholic fatty liver disease. *Hepatology* **58**, 120–127 (2013).
80. P. Chen, Q. Luo, C. Huang, Q. Gao, L. Li, J. Chen, B. Chen, W. Liu, W. Zeng, Z. Chen, Pathogenesis of non-alcoholic fatty liver disease mediated by YAP. *Hepatol. Int.* **12**, 26–36 (2018).
81. H. Zhang, H. A. Pasolli, E. Fuchs, Yes-associated protein (YAP) transcriptional coactivator functions in balancing growth and differentiation in skin. *Proc. Natl. Acad. Sci. U.S.A.* **108**, 2270–2275 (2011).
82. M. J. Lee, M. R. Byun, M. Furutani-Seiki, J. H. Hong, H. S. Jung, YAP and TAZ regulate skin wound healing. *J. Invest. Dermatol.* **134**, 518–525 (2014).
83. J. M. Goffin, P. Pittet, G. Csucs, J. W. Lussi, J.-J. Meister, B. Hinz, Focal adhesion size controls tension-dependent recruitment of α-smooth muscle actin to stress fibers. *J. Cell Biol.* **172**, 259–268 (2006).
84. S. Mascharak, H. E. desJardins-Park, M. F. Davitt, M. Griffin, M. R. Borrelli, A. L. Moore, K. Chen, B. Duoto, M. Chinta, D. S. Foster, A. H. Shen, M. Januszyk, S. H. Kwon, G. Wernig, D. C. Wan, H. P. Lorenz, G. C. Gurtner, M. T. Longaker, Preventing *Engrailed-1* activation in fibroblasts yields wound regeneration without scarring. *Science* **372**, eaba2374 (2021).
85. D. C. Stewart, D. Berrie, J. Li, X. Liu, C. Rickerson, D. Mkoji, A. Iqbal, S. Tan, A. L. Doty, S. C. Glover, C. S. Simmons, Quantitative assessment of intestinal stiffness and associations with fibrosis in human inflammatory bowel disease. *PLOS ONE* **13**, e0200377 (2018).
86. S. Yui, L. Azzolin, M. Maimets, M. T. Pedersen, R. P. Fordham, S. L. Hansen, H. L. Larsen, J. Guiu, M. R. P. Alves, C. F. Rundsten, J. V. Johansen, Y. Li, C. D. Madsen, T. Nakamura, M. Watanabe, O. H. Nielsen, P. J. Schweiger, S. Piccolo, K. B. Jensen, YAP/TAZ-dependent reprogramming of colonic epithelium links ECM remodeling to tissue regeneration. *Cell Stem Cell* **22**, 35–49.e7 (2018).
87. O. Guillermin, N. Angelis, C. M. Sidor, R. Ridgway, A. Baulies, A. Kucharska, P. Antas, M. R. Rose, J. Cordero, O. Sansom, V. S. W. Li, B. J. Thompson, Wnt and Src signals converge on YAP-TEAD to drive intestinal regeneration. *EMBO J.* **40**, e105770 (2021).
88. J. Cai, N. Zhang, Y. Zheng, R. F. de Wilde, A. Maitra, D. Pan, The Hippo signaling pathway restricts the oncogenic potential of an intestinal regeneration program. *Genes Dev.* **24**, 2383–2388 (2010).
89. A. von Gise, Z. Lin, K. Schlegelmilch, L. B. Honor, G. M. Pan, J. N. Buck, Q. Ma, T. Ishiwata, B. Zhou, F. D. Camargo, W. T. Pu, YAP1, the nuclear target of Hippo signaling, stimulates heart growth through cardiomyocyte proliferation but not hypertrophy. *Proc. Natl. Acad. Sci. U.S.A.* **109**, 2394–2399 (2012).
90. M. Xin, Y. Kim, L. B. Sutherland, M. Murakami, X. Qi, J. McAnally, E. R. Porrello, A. I. Mahmoud, W. Tan, J. M. Shelton, J. A. Richardson, H. A. Sadek, R. Bassel-Duby,

E. N. Olson, Hippo pathway effector Yap promotes cardiac regeneration. *Proc. Natl. Acad. Sci. U.S.A.* **110**, 13839–13844 (2013).
91. M. F. Berry, A. J. Engler, Y. J. Woo, T. J. Pirolli, L. T. Bish, V. Jayasankar, K. J. Morine, T. J. Gardner, D. E. Discher, H. L. Sweeney, Mesenchymal stem cell injection after myocardial infarction improves myocardial compliance. *Am. J. Physiol. Heart Circ. Physiol.* **290**, H2196–H2203 (2006).
92. Z. Lin, A. von Gise, P. Zhou, F. Gu, Q. Ma, J. Jiang, A. L. Yau, J. N. Buck, K. A. Gouin, P. R. van Gorp, B. Zhou, J. Chen, J. G. Seidman, D. Z. Wang, W. T. Pu, Cardiac-specific YAP activation improves cardiac function and survival in an experimental murine MI model. *Circ. Res.* **115**, 354–363 (2014).
93. D. Shao, P. Zhai, D. P. Del Re, S. Sciarretta, N. Yabuta, H. Nojima, D. S. Lim, D. Pan, J. Sadoshima, A functional interaction between Hippo-YAP signalling and FoxO1 mediates the oxidative stress response. *Nat. Commun.* **5**, 3315 (2014).
94. M. M. Mia, D. M. Cibi, S. A. Binte Abdul Ghani, A. Singh, N. Tee, V. Sivakumar, H. Bogireddi, S. A. Cook, J. Mao, M. K. Singh, Loss of Yap/taz in cardiac fibroblasts attenuates adverse remodeling and improves cardiac function. *Cardiovasc. Res.* **118**, 1785–1804 (2021).
95. S. Kurotsu, T. Sadahiro, R. Fujita, H. Tani, H. Yamakawa, F. Tamura, M. Isomi, H. Kojima, Y. Yamada, Y. Abe, Y. Murakata, T. Akiyama, N. Muraoka, I. Harada, T. Suzuki, K. Fukuda, M. Ieda, Soft matrix promotes cardiac reprogramming via inhibition of YAP/TAZ and suppression of fibroblast signatures. *Stem Cell Rep.* **15**, 612–628 (2020).
96. T. Sun, Z. Huang, H. Zhang, C. Posner, G. Jia, T. R. Ramalingam, M. Xu, H. Brightbill, J. G. Egen, A. Dey, J. R. Arron, TAZ is required for lung alveolar epithelial cell differentiation after injury. *JCI Insight* **5**, e128674 (2019).
97. Z. Liu, H. Wu, K. Jiang, Y. Wang, W. Zhang, Q. Chu, J. Li, H. Huang, T. Cai, H. Ji, C. Yang, N. Tang, MAPK-mediated YAP activation controls mechanical-tension-induced pulmonary alveolar regeneration. *Cell Rep.* **16**, 1810–1819 (2016).
98. M. Sun, Y. Sun, Z. Feng, X. Kang, W. Yang, Y. Wang, Y. Luo, New insights into the Hippo/YAP pathway in idiopathic pulmonary fibrosis. *Pharmacol. Res.* **169**, 105635 (2021).
99. S. Noguchi, A. Saito, Y. Mikami, H. Urushiyama, M. Horie, H. Matsuzaki, H. Takeshima, K. Makita, N. Miyashita, A. Mitani, T. Jo, Y. Yamauchi, Y. Terasaki, T. Nagase, TAZ contributes to pulmonary fibrosis by activating profibrotic functions of lung fibroblasts. *Sci. Rep.* **7**, 42595 (2017).
100. F. Liu, D. Lagares, K. M. Choi, L. Stopfer, A. Marinkovic, V. Vrbanac, C. K. Probst, S. E. Hiemer, T. H. Sisson, J. C. Horowitz, I. O. Rosas, L. E. Fredenburgh, C. Feghali-Bostwick, X. Varelas, A. M. Tager, D. J. Tschumperlin, Mechanosignaling through YAP and TAZ drives fibroblast activation and fibrosis. *Am. J. Physiol. Lung Cell. Mol. Physiol.* **308**, L344–L357 (2015).
101. B. Hinz, Mechanical aspects of lung fibrosis. *Proc. Am. Thorac. Soc.* **9**, 137–147 (2012).
102. Y. Liu-Chittenden, B. Huang, J. S. Shim, Q. Chen, S. J. Lee, R. A. Anders, J. O. Liu, D. Pan, Genetic and pharmacological disruption of the TEAD-YAP complex suppresses the oncogenic activity of YAP. *Genes Dev.* **26**, 1300–1305 (2012).
103. V. S. Athwal, J. Pritchett, J. Llewellyn, K. Martin, E. Camacho, S. M. Raza, A. Phythian-Adams, L. J. Birchall, A. F. Mullan, K. Su, L. Pearmain, G. Dolman, A. M. Zaitoun, S. L. Friedman, A. MacDonald, W. L. Irving, I. N. Guha, N. A. Hanley, K. Piper Hanley, SOX9 predicts progression toward cirrhosis in patients while its loss protects against liver fibrosis. *EMBO Mol. Med.* **9**, 1696–1710 (2017).
104. J. J. Gokey, A. Sridharan, Y. Xu, J. Green, G. Carraro, B. R. Stripp, A. T. Perl, J. A. Whitsett, Active epithelial Hippo signaling in idiopathic pulmonary fibrosis. *JCI Insight* **3**, e98738 (2018).
105. T. Toyama, A. P. Looney, B. M. Baker, L. Stawski, P. Haines, R. Simms, A. D. Szymaniak, X. Varelas, M. Trojanowska, Therapeutic targeting of TAZ and YAP by dimethyl fumarate in systemic sclerosis fibrosis. *J. Invest. Dermatol.* **138**, 78–88 (2018).
106. G. Sorrentino, N. Ruggeri, V. Specchia, M. Cordenonsi, M. Mano, S. Dupont, A. Manfrin, E. Ingallina, R. Sommaggio, S. Piazza, A. Rosato, S. Piccolo, G. Del Sal, Metabolic control of YAP and TAZ by the mevalonate pathway. *Nat. Cell Biol.* **16**, 357–366 (2014).
107. C. Y. Wang, P. Y. Liu, J. K. Liao, Pleiotropic effects of statin therapy: Molecular mechanisms and clinical results. *Trends Mol. Med.* **14**, 37–44 (2008).
108. S. Jiao, H. Wang, Z. Shi, A. Dong, W. Zhang, X. Song, F. He, Y. Wang, Z. Zhang, W. Wang, X. Wang, T. Guo, P. Li, Y. Zhao, H. Ji, L. Zhang, Z. Zhou, A peptide mimicking VGLL4 function acts as a YAP antagonist therapy against gastric cancer. *Cancer Cell* **25**, 166–180 (2014).
109. L. Chang, L. Azzolin, D. Di Biagio, F. Zanconato, G. Battilana, R. Lucon Xiccato, M. Aragona, S. Giulitti, T. Panciera, A. Gandin, G. Sigismondo, J. Krijgsveld, M. Fassan, G. Brusatin, M. Cordenonsi, S. Piccolo, The SWI/SNF complex is a mechanoregulated inhibitor of YAP and TAZ. *Nature* **563**, 265–269 (2018).
110. J. Yong, Y. Li, S. Lin, Z. Wang, Y. Xu, Inhibitors targeting YAP in gastric cancer: Current status and future perspectives. *Drug Des. Devel. Ther.* **15**, 2445–2456 (2021).
111. A. Srivastava, In vivo tissue-tropism of adeno-associated viral vectors. *Curr. Opin. Virol.* **21**, 75–80 (2016).
112. L. Liu, S. X. Zhang, W. Liao, H. P. Farhoodi, C. W. Wong, C. C. Chen, A. I. Segaliny, J. V. Chacko, L. P. Nguyen, M. Lu, G. Polovin, E. J. Pone, T. L. Downing, D. A. Lawson, M. A. Digman, W. Zhao, Mechanoresponsive stem cells to target cancer metastases through biophysical cues. *Sci. Transl. Med.* **9**, eaan2966 (2017).
113. P. K. Veerasubramanian, A. Trinh, N. Akhtar, W. F. Liu, T. L. Downing, Biophysical and epigenetic regulation of cancer stemness, invasiveness, and immune action. *Curr. Tissue Microenviron. Rep.* **1**, 277–300 (2020).
114. T. Wang, T. U. Luu, A. Chen, M. Khine, W. F. Liu, Topographical modulation of macrophage phenotype by shrink-film multi-scale wrinkles. *Biomater. Sci.* **4**, 948–952 (2016).
115. T. U. Luu, S. C. Gott, B. W. Woo, M. P. Rao, W. F. Liu, Micro- and nanopatterned topographical cues for regulating macrophage cell shape and phenotype. *ACS Appl. Mater. Interfaces* **7**, 28665–28672 (2015).
116. P. K. Veerasubramanian, V. C. Joe, W. F. Liu, T. L. Downing, Characterization of macrophage and cytokine interactions with biomaterials used in negative-pressure wound therapy. *Bioengineering* **9**, 2 (2021).
117. F. Y. McWhorter, T. Wang, P. Nguyen, T. Chung, W. F. Liu, Modulation of macrophage phenotype by cell shape. *Proc. Natl. Acad. Sci. U.S.A.* **110**, 17253–17258 (2013).
118. R. G. Scheraga, S. Abraham, K. A. Niese, B. D. Southern, L. M. Grove, R. D. Hite, C. McDonald, T. A. Hamilton, M. A. Olman, TRPV4 mechanosensitive ion channel regulates lipopolysaccharide-stimulated macrophage phagocytosis. *J. Immunol.* **196**, 428–436 (2016).
119. H. Atcha, A. Jairaman, J. R. Holt, V. S. Meli, R. R. Nagalla, P. K. Veerasubramanian, K. T. Brumm, H. E. Lim, S. Othy, M. D. Cahalan, M. M. Pathak, W. F. Liu, Mechanically activated ion channel Piezo1 modulates macrophage polarization and stiffness sensing. *Nat. Commun.* **12**, 3256 (2021).
120. R. Sridharan, B. Cavanagh, A. R. Cameron, D. J. Kelly, F. J. O'Brien, Material stiffness influences the polarization state, function and migration mode of macrophages. *Acta Biomater.* **89**, 47–59 (2019).
121. A. K. Blakney, M. D. Swartzlander, S. J. Bryant, The effects of substrate stiffness on the in vitro activation of macrophages and in vivo host response to poly(ethylene glycol)-based hydrogels. *J. Biomed. Mater. Res. A* **100**, 1375–1386 (2012).
122. T. Okamoto, Y. Takagi, E. Kawamoto, E. J. Park, H. Usuda, K. Wada, M. Shimaoka, Reduced substrate stiffness promotes M2-like macrophage activation and enhances peroxisome proliferator-activated receptor γ expression. *Exp. Cell Res.* **367**, 264–273 (2018).
123. V. S. Meli, P. K. Veerasubramanian, H. Atcha, Z. Reitz, T. L. Downing, W. F. Liu, Biophysical regulation of macrophages in health and disease. *J. Leukoc. Biol.* **106**, 283–299 (2019).
124. J. C. Escolano, A. V. Taubenberger, S. Abuhattum, C. Schweitzer, A. Farrukh, A. Del Campo, C. E. Bryant, J. Guck, Compliant substrates enhance macrophage cytokine release and NLRP3 inflammasome formation during their pro-inflammatory response. *Front. Cell Dev. Biol.* **9**, 639815 (2021).
125. E. Gruber, C. Heyward, J. Cameron, C. Leifer, Toll-like receptor signaling in macrophages is regulated by extracellular substrate stiffness and Rho-associated coiled-coil kinase (ROCK1/2). *Int. Immunol.* **30**, 267–278 (2018).
126. H. Joshi, A. Almgren-Bell, E. P. Anaya, E. M. Todd, S. J. Van Dyken, A. Seth, K. M. McIntire, S. Singamaneni, F. Sutterwala, S. C. Morley, L-plastin enhances NLRP3 inflammasome assembly and bleomycin-induced lung fibrosis. *Cell Rep.* **38**, 110507 (2022).
127. H. J. Choi, N. E. Kim, B. M. Kim, M. Seo, J. H. Heo, TNF-α-induced YAP/TAZ activity mediates leukocyte-endothelial adhesion by regulating VCAM1 expression in endothelial cells. *Int. J. Mol. Sci.* **19**, 3428 (2018).
128. Y. Lv, K. Kim, Y. Sheng, J. Cho, Z. Qian, Y. Y. Zhao, G. Hu, D. Pan, A. B. Malik, G. Hu, YAP controls endothelial activation and vascular inflammation through TRAF6. *Circ. Res.* **123**, 43–56 (2018).
129. R. Caire, E. Dalix, M. Chafchafi, M. Thomas, M. T. Linossier, M. Normand, A. Guignandon, L. Vico, H. Marotte, YAP transcriptional activity dictates cell response to TNF in vitro. *Front. Immunol.* **13**, 856247 (2022).

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