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Review

The Hippo Pathway, YAP/TAZ, and the Plasma Membrane

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The plasma membrane allows the cell to sense and adapt to changes in the extracellular environment by relaying external inputs via intracellular signaling networks. One central cellular signaling pathway is the Hippo pathway, which regulates homeostasis and plays chief roles in carcinogenesis and regenerative processes. Recent studies have found that mechanical stimuli and diffusible chemical components can regulate the Hippo pathway primarily through receptors embedded in the plasma membrane. Morphologically defined structures within the plasma membrane, such as cellular junctions, focal adhesions, primary cilia, caveolae, clathrin-coated pits, and plaques play additional key roles. Here, we discuss recent evidence highlighting the importance of these specialized plasma membrane domains in cellular feedback via the Hippo pathway.

Highlights

Plasma membrane structures direct dynamic and context-specific cellular signaling via the Hippo pathway.

Hippo pathway components 'moonlight' via interactions with plasma membrane structures.

YAZ and TAZ drive expression of components involved in the generation of specialized plasma membrane domains.

The Hippo pathway is integrated into multiple levels of cellular feedback.

Cellular Regulation by the Hippo Pathway

The plasma membrane is essential for cell integrity and serves as an interface to sense and respond to changes in the extracellular environment [1]. A large variety of plasma membrane domains, such as, **adherens and tight junctions** (see Glossary), **focal adhesions (FAs)**, **clathrin-coated pits (CCPs)** or plaques, **caveolae**, and **primary cilia** [2–8], allow the cell to dynamically relay chemical and mechanical stimuli, which are translated into direct cellular responses. The plasma membrane as a whole, but FAs in particular, strongly interacts with the **extracellular matrix (ECM)** [9]. The ECM is a dynamic noncellular matrix surrounding cells and tissues that acts as a scaffold for cell anchorage and mechanotransduction [9]. The signals perceived by plasma membrane elements are integrated and transmitted by a variety of signaling pathways. One central pathway, which enables the cell to respond to various signals, is the Hippo pathway (Box 1) [10–13]. By highly context specific responses the Hippo pathway regulates cellular homeostasis and plays central roles in carcinogenesis and regenerative processes [10,12,13]. The Hippo pathway is extracellularly regulated by mechanical stimuli and diffusible chemicals. These signals are sensed in great part by receptors, such as G-protein coupled receptors (GPCRs) and adherence complexes embedded in the plasma membrane [1,10–16]. To ensure a highly specific response, junctional complexes and receptors accumulate in distinct membrane structures and their plasma membrane abundance is furthermore dynamically regulated by exo- and endocytosis [3,4,7,17]. Junctional complexes provide robust cellular sensitivity of Hippo signaling to cell polarity and cell–cell contacts [10,15,16]. Catenins [18,19], protein tyrosine phosphatase nonreceptor (PTPN)14 [20,21], and the angiomotin family [22–26] play central roles in this regulation as direct YAP-binding proteins. Both PTPN14 and AMOT interact via PPxY motifs with WW domains of YAP and TAZ, and consequently, this interaction does not directly require YAP and TAZ Hippo-pathway-mediated phosphorylation [20–23]. Several of the Hippo pathway components temporally localize to junctional complexes, including YAP and TAZ, KIBRA, LATS1/2, neurofibromatosis type 2 (NF2), and MST1/2 [24,26–29]. At the junctional location the upstream Hippo pathway components are activated, and consequently, YAP/TAZ are inhibited. As cellular junctions function as mechanical cellular transducers, this spatiotemporal localization of Hippo pathway components brings them proximal to sense the exerted forces. The interplay between cellular junctions and the Hippo pathway is well established [10,15,16]. Here, we discuss recent evidence highlighting that additional plasma membrane domains, such as FAs, CCPs and plaques, caveolae, and primary cilia provide cellular feedback via the Hippo pathway.

The Hippo Pathway, YAP/TAZ, Integrins, and FAs

Clustering of **integrins** increases the avidity of the multivariant interactions with the ECM substrate [30,31]. FAs are large dynamic multiprotein complexes comprised of several distinct layers of proteins with the integral components integrins, vinculin, talin, and focal adhesion kinase (FAK) [6], and

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Box 1. The Hippo Pathway and Regulation of YAP/TAZ

The core Hippo pathway signaling cascade in mammals is comprised of a serine/threonine kinase cascade consisting of MST1/2 (homologs of the *Drosophila* kinase Hippo), interacting with the scaffolding proteins Salvador homolog 1 (SAV1) and neurofibromatosis type 2 (NF2/Merlin), as well as LATS1/2, which interact with MOB kinase activator 1A and B (MOB1A and B) [10,16,154,155] (Figure I). In the canonical Hippo pathway (components highlighted in magenta), MST1/2 interact with SAV1 and phosphorylate LATS1/2, which are activated and phosphorylate YAP/TAZ on five (YAP) and four (TAZ) conserved serine residues. These inhibitory phosphorylations of YAP and its paralog TAZ is a signal for the cytoplasmic retention and YAP/TAZ binding to 14-3-3 protein or YAP/TAZ degradation [10,154]. This activation of MST1/2 and LATS1/2 denotes the Hippo pathway on state, where YAP/TAZ are inactive. In addition, Hippo (MST1/2)-independent, LATS1/2-mediated regulation of YAP/TAZ also occurs via the MAP4K kinase family [154], as well as by STK25 [156]. Under certain circumstances the nuclear dbf2-related1/2 kinases (NDR1/2), substrates of MAP4K, MST1/2, and STK24 (MST3), directly phosphorylate and inhibit YAP [157–161]. This additional network of kinases (highlighted in blue in Figure I) provides additional means for signal input, cellular adaptability, and robustness. Unphosphorylated YAP/TAZ translocate into the nucleus where they primarily interact with TEAD1–4 to regulate gene transcription [13,49,59,162]. The activity of the Hippo pathway core kinases is regulated by various stimuli; for example, cell–cell contact, extracellular signals, cell polarity, metabolic state, and mechanotransduction [10,11,33,59]. In addition, SRC-activating phosphorylation of YAP and SRC-inhibitory phosphorylation of LATS facilitate YAP nuclear localization and induction of gene transcription [32,36,61,163,164]. Additional kinase mediated regulation of YAP/TAZ via NLK [119,120], 5′ AMP-activated protein kinase (AMPK), cyclin-dependent kinase (CDK), and others are also incorporated [13,16,154]. The quality of the integrated signals leads either to activation or inhibition of the cotranscriptional activators YAP and TAZ, and allows a specific and timely regulation of gene transcription [10,11,13,16,162].

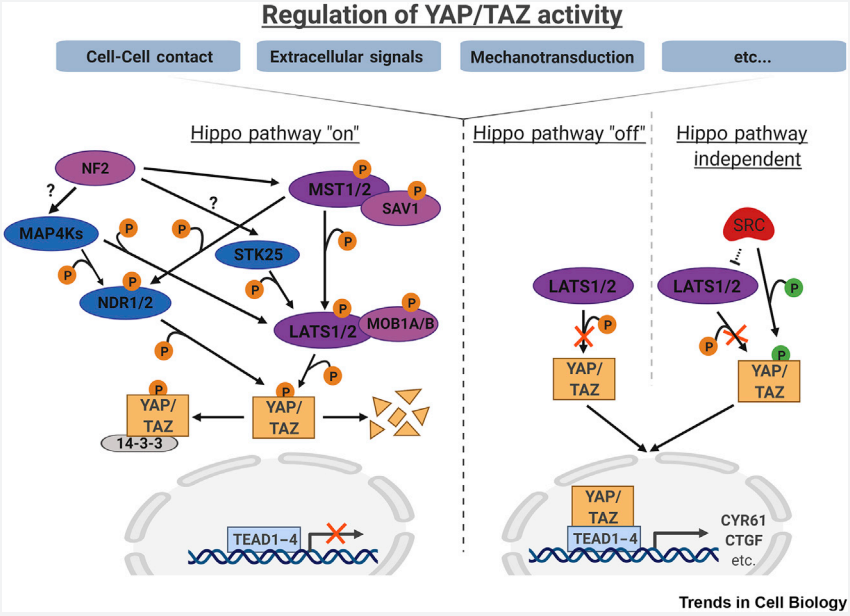

Figure I. The Hippo Pathway and Regulation of YAP/TAZ.

thereby relay mechanical responses from large integrin complexes at the plasma membrane. FAs thereby provide mechanical links between the intracellular cytoskeleton and the ECM. These protein complexes function as mechanosensors and -integrators and coordinate a diverse array of signaling molecules [6], including the Hippo pathway [32].

Integrins, via the tyrosine kinase SRC, as well as FAK activate YAP [32–37]. Increased ECM stiffness, relayed especially via β1 integrins, activates FAK, which in turn activates the tyrosine kinase SRC.

Glossary

Adherens and tight junctions: distinct cell–cell transmembrane protein complexes that provide adhesive contacts between neighboring cells [8,15]. The extracellular adhesive contacts between cells are directly linked to the intracellular actin cytoskeleton. A range of intracellular signaling molecules, such as catenins (adherens junctions) and zonula occludens (ZO) proteins (tight junctions) relay the cellular information. In vertebrate epithelial cells, tight junctions localize just apical to adherens junctions. Tight junctions contribute to the establishment and maintenance of the apical–basal polarity. Both types of junctions are regulators of gene transcription and major regulators of many signaling pathways, including the Hippo pathway [8,15].

Caveolae: 50–80-nm plasma membrane invaginations [166,167] involved in a variety of cellular tasks: endocytosis, maintenance of the membrane lipid composition, metabolism, cell signaling, and mechanosensing/-protection [2,5,17,96].

Clathrin-coated pits (CCPs): 80–100-nm clathrin-coated invaginations at the plasma membrane, including a range of adaptor proteins, which accumulate receptors and are able to pinch off, in a highly regulated and dynamin dependent manner, in a process termed clathrin-mediated endocytosis [80]. CCPs perform selective vesicular uptake of cargo [4]. The molecular building blocks of CCPs are the clathrin triskelions [184], each composed of three clathrin heavy chains and three light chains [4,165]. The triskila form the polyhedral lattice coat that makes up the characteristic clathrin coat [75,165,184].

Clathrin-coated plaques/structures/lattices (CCSs): unlike CCPs, plaques are patches of clathrin accumulated at the plasma membrane. CCSs have diverse functions, ranging from endocytosis to cell adhesion and mechanotransduction. In some instances, CCPs form at the rim of plaques [3,75,84].

Desmosomes: (also known as *maculae adherents*) are widely expressed and composed of the

SRC subsequently phosphorylates and activates YAP while also phosphorylating and inhibiting LATS1/2. Consequently, this activates YAP via two separate means (Figure 1A, Box 1) [32,35–37]. Conversely, upon FAK inhibition, YAP nuclear localization and activity are reduced even upon fibronectin stimulation (Figure 1B) [32]. PAK-family kinases are serine/threonine kinases that regulate FA dynamics and also inhibit LATS-mediated YAP phosphorylation. FAK activates PAK1 via AKT serine/threonine kinase 1 or the small GTPases, CDC42 or RAC. PAK1 phosphorylates and inactivates the upstream Hippo kinase activator NF2. As a result, LATS-mediated inhibitory YAP phosphorylation is downregulated (Figure 1C) [36,38,39]. RAP2 (Ras-related GTPase) facilitates integrin-mediated cell adhesion [40]. RAP2 is an upstream regulator of the Hippo pathway in response to mechanical stimuli and transmits changes in ECM stiffness to the cell and regulates YAP/TAZ activity [41]. At low ECM rigidity, RAP2 activates Rho GTPase-activating protein (RhoGAP), ARHGAP29, and MAP4Ks, which leads to LATS1/2 activation and inhibitory phosphorylation of YAP/TAZ (Figure 1D) [41–43]. Consequently, upon RAP2 deletion YAP/TAZ remain active, even at low ECM stiffness [41]. Rho-GTPases, integral parts of FA assembly and stability, activate YAP and TAZ [34,44,45]. The p190 RhoGAPs p190A and p190B are RhoA GAPs and predominantly function through RhoA inactivation. Loss-of-function mutations of p190A, encoded by *ARHGAP35*, are a frequent occurrence in cancers such as uterine, bladder, stomach, and lung cancers. Depletion of epithelial p190A and/or p190B (encoded by *ARHGAP5*) leads to increased RhoA activity [46,47], which causes YAP activation [48]. The epithelial p190A/p190B-deficient cells do not have any gross defects in adherens or tight junction formation, but instead have increased FAs [48]. Both p190A and p190B are repressors of contact inhibition of proliferation (CIP) [47,48]. Likewise, YAP/TAZ activity is repressed upon cell–cell contact [18,49]. p190 RhoGAPs inhibit the Rho–ROCK pathway, which activates LATS1/2 and represses YAP activity and consequently promotes CIP [48].

Upon mechanical stress, the PDZ and LIM domain containing Enigma proteins bind to α-actinin and actin stress fibers at FAs. Enigma proteins appear to serve as binding platforms for YAP. Association of YAP with this complex facilitates the SRC-mediated activating tyrosine phosphorylation. This protein complex enables YAP nuclear translocation and consequently YAP-mediated gene transcription [50]. The combined cellular depletion of the two Enigma proteins (encoded by *PDLIM5* and *PDLIM7*) renders YAP cytosolic and intriguingly even appears to override the Hippo-pathway-mediated regulation of YAP [50].

Additional levels of mechanoregulation of YAP occurs directly at the nuclear pore, some of which might be FA mediated. The nucleus is elongated upon increased substrate stiffness and tensile stresses, which, via the linker of nucleoskeleton and cytoskeleton (LINC) complex [51], provides a three-way feedback between the adhesions, cytoskeleton, and nucleus [52,53]. This response originates from ECM–FA interactions and is relayed via stress fibers that stretch the nucleus [52,53], resulting in the widening of the nuclear pore, allowing YAP to enter the nucleus and activate gene transcription [54]. However, the nucleus is itself mechanosensitive, as exerting tension on isolated nuclei induces stiffening of the nucleus [55], and stretching of the nuclear membrane directly leads to YAP nuclear translocation [54] implying that FAs, under certain circumstances, are not needed for this nuclear-mediated YAP filtering. It is still unclear how the nuclear pore retains selectivity upon stretching, and also what role the Enigma proteins (if any) might play in this process [54].

Remodeling of the actin cytoskeleton directly affects FA dynamics and is also a key mediator of YAP/TAZ activity [33,34,37]. In both mammals and zebrafish, YAP/TAZ activation regulates RhoGAPs, which consequently remodels the cytoskeleton, and this in return affects the initiation and maintenance of FAs [32,33,56]. YAP/TAZ induction of *ARHGAP18* causes increased cytoskeletal tension [57], while YAP induced activation, in a cell-type-dependent manner, of either *ARHGAP28* or *ARHGAP29*, results in F-actin destabilization [37,58,59]. Besides interaction with the cytoskeleton, regulation of FA components via YAP/TAZ is key for FA integrity and dynamics [37,56,60,61]. Expression of distinct integrins (e.g., αVβ1 and αVβ3) and FA components are directly or indirectly regulated by YAP/TAZ–TEAD. Loss of YAP/TAZ–TEAD activity therefore dramatically changes the overall cellular composition of integrins [56,59]. Furthermore, loss of YAP induces disruption of integrin subunit interaction [56]. In YAP-deficient cells integrin heterodimer formation is reduced and

single transmembrane proteins desmogleins (Dsgs) and desmocollins (Dscs), as well as the cytoplasmic localized tarmadillo family proteins, plakoglobin (γ-catenin), and the plakophilins (Pkp1–3). Desmosomes tether intermediate filaments to the plasma membrane and neighboring cells and are essential for stable intercellular cohesion [148].

Extracellular matrix (ECM): noncellular matrix present in all tissues and central for cell homeostasis. The composition of the ECM includes proteoglycans and fibrous proteins (e.g., collagens, laminins, fibronectins, and elastins). The ECM provides a complex physical 3D scaffold, that functions as a cellular anchor. Responding to mechanical and chemical stimuli, the ECM is actively remodeled and mediate mechanical stresses within and between cells and tissues [9].

Focal adhesions (FAs): clusters of proteins; mainly integrins, FAKs, talins, vinculins, paxilins, etc. FAs are located in the plasma membrane and interact with the ECM and the cytoskeleton. They provide an interface with the ECM and transmit mechanical stimuli through multiple cellular signaling pathways [6].

Integrins: heterodimeric transmembrane adhesion receptors composed of α and β subunits, forming 24 different mammalian integrins, which differ in their binding capacity and cell-type-dependent expression [30,31]. The adhesion receptors are central in several types of plasma membrane complexes, FAs, and clathrin-coated structures. Integrins bind ECM fibrils while the intracellular portion interacts with the cytoskeleton. Integrins can be bidirectionally activated by receiving signals from the ECM as well as the cytoskeleton [30,31].

Primary cilia: plasma membrane organelle formed during growth arrest. It is nonmotile and comprises a set structure of microtubules [7,135]. Due to the dynamic localization of specific receptors, ion channels, and transporters, this membrane evagination has a central role in cell signaling. Consequently, primary cilia serves as a sensory cellular extension that coordinates key cellular functions [7,135,142].

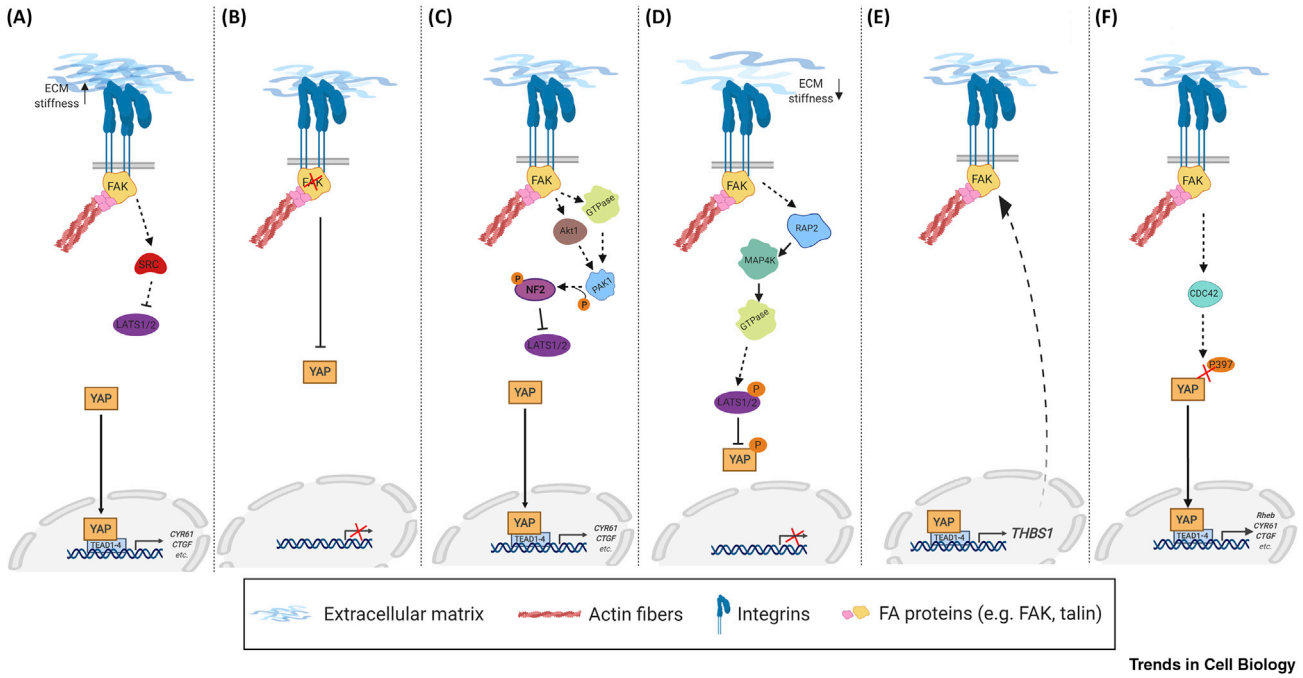

Figure 1. Hippo Pathway and Focal Adhesions (FAs).

FAs are mechanotransducing hubs that integrate and relay mechanical cues arising from the extracellular milieu to the cellular cytoskeleton. Interactions between the Hippo pathway components YAP and TAZ and FAs are multitude and mediated via focal adhesion kinase (FAK) and frequently constitute a feedforward loop. (A) Increased extracellular matrix (ECM) stiffness is sensed by FAs, which activate SRC and inhibit LATS1/2, and consequently, YAP is activated. (B) FAK inhibition causes YAP inactivation. (C) FAs activation prompt PAK1 activation that phosphorylates (and thereby inhibits) NF2, which causes inactivation of LATS1/2, and consequently, YAP is activated. (D) A decrease in ECM stiffness is sensed via FAs and relayed via the GTPase RAP2, which bind to and stimulate the MAP4Ks causing LATS1/2 activation and YAP inhibition. (E) YAP–TEAD induces THBS1 that engages FAs and activate FAK. (F) FAs, via FAK and through CDC42, decrease the LATS-mediated inhibitory Ser397 phosphorylation of YAP.

consequently the number of FAs is decreased [56,61]. Moreover, YAP deficiency leads to decreased transcription of genes encoding FA components (e.g., vinculin and zyxin) as well as diminished S157 phosphorylation of the actin docking protein (VASP); a site that is required for VASP localization at FAs. As a consequence, the interaction between FAs and the cytoskeleton is disrupted and subsequently reduces FAs in YAP-deficient cells [56]. YAP–TEAD in breast cancer cells directly induces expression of the adhesive matrix glycoprotein thrombospondin (THBS)1, which activates FAK [60] (Figure 1E). In addition, YAP-induced ECM components, such as collagens and fibronectin, provide an additional feed-forward loop that activates FAK [41,56,61]. YAP is required for a specialized type of directed cell migration termed durotaxis [35]. During durotaxis, anisotropic mechanical stimulation prompts directed motility through FA and the actomyosin cytoskeleton [41,56,61], and cells respond to a gradient of extracellular stiffness by migrating toward increasing matrix stiffness [35,62,63]. FAs mediate via dynamic actin polymerization an oscillating traction force, which mechanically directs this motility, and both functional FAK as well as YAP are essential [35,62–64]. Durotaxis has wide implications in both development and cancer [35,62–64]. FAK activation in return promotes FA stability and tumor invasiveness [60]. In flies, Yki (YAP ortholog) induces Stretchin–Mlck-mediated myosin activation, which leads to cellular tension and promotes cell growth [65]. An example of the importance of the complex and integrated FA–YAP/TAZ–TEAD cellular interplay is that in stem cells of the mouse incisor an integrin α3–FAK–CDC42 signaling axis leads to a decrease in the LATS-mediated inhibitory S397 YAP phosphorylation [66] (Figure 1F). As a result, YAP translocates to the nucleus and induces *Rheb* expression and consequently activates mTOR signaling [66]. Integration of mTOR with the Hippo pathway via YAP/TAZ–TEAD-regulated gene induction of prominent mTOR activators is a widespread phenomenon [67–69], and through this it regulates metabolism, cell competition, and cell

size [67–69]. YAP/TAZ–TEAD drives expression of the heterodimeric disulfide-linked plasma membrane resident CD98 (encoded by *SLC7A5* and *SLC3A2*) [67,69]. CD98, also known as LAT1, is an amino acid transporter that transports essential amino acids, such as leucine, into the cell, which activates mTOR [70], but CD98 also functions as a mediator of integrin-dependent adhesion [70–73]. CD98 is frequently overexpressed in cancer and increased expression of CD98 activates YAP/TAZ, providing a feed-forward loop [67,69,72]. This integration with mTOR might explain how integrin α3β1 increases proliferation and sustained tumor growth in skin cancer, and further indicates the complexity of YAP/TAZ and FA interactions, that influence cell integrity and cell fate [66,74].

Overall, FAs and integrins are key mechanosensory elements of the cell and regulate YAP activity in response to mechanical cues [32,35–37]. The interplay and integrated feedback mechanism between the ECM, integrins, FA, cytoskeleton, and Hippo pathway on multiple levels provide a robust way for cells to differentially respond to dynamic changes.

The Hippo Pathway, YAP/TAZ, Clathrin-Coated Pits, and Clathrin-Coated Structures/Plaques/Lattices

CCPs (Box 2) and **clathrin-coated structures/plaques/lattices (CCSs)** are specialized areas in the plasma membrane with accumulations of clathrin, which are primed for selective endocytosis, but also function as adhesion complexes [3,4,75,76]. CCPs and CCSs interact closely with the actin

Box 2. Clathrin-Coated Pits

CCPs transiently assemble at the plasma membrane and are primed to pinch off the membrane in a dynamin-dependent and highly coordinated process termed clathrin-dependent endocytosis [3] (Figure II). This endocytic process is a strictly regulated and efficient process that selects and concentrates cargo molecules (such as receptors and ligand bound receptors); these specific extracellular constituents are then internalized [3,4,75,80,165]. Clathrin-dependent endocytosis (CDE) is fast (the coordinated process from initiation of invagination to fully pinched off vesicles frequently occurs within minutes) [76,80]. CDE is an adaptable process, as sorting into CDE, the rate of CDE and intracellular sorting of cargo is regulated by cellular stimuli [4,75,80,165]. CDE therefore plays major roles in cellular metabolism, and the ability of cells to respond to and differentiate between various cellular stimuli. CDE is a prominent way for cells to downregulate cell surface receptors, but CDE is also important in absorption of essential nutrients, such as by the uptake of the well-established CDE ligand transferrin, and the activation of intracellular signal transduction cascades [4,75,80,165].

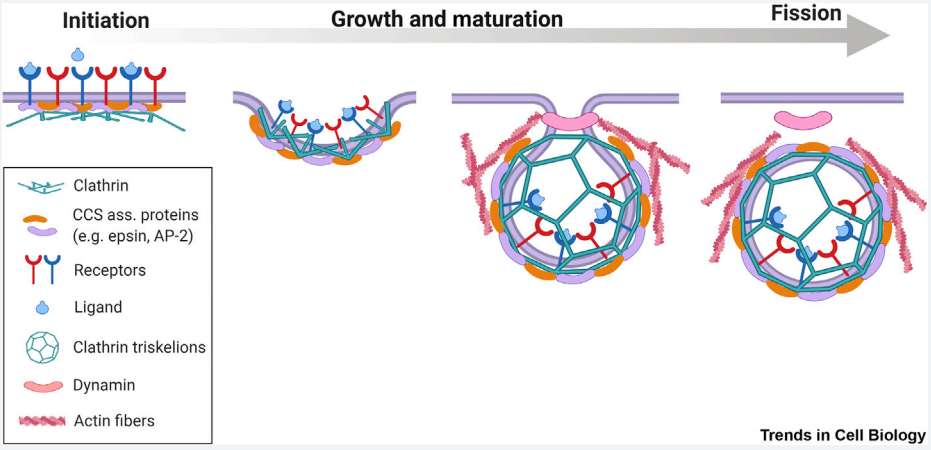

Figure II. Clathrin-Coated Pits.

cytoskeleton [77–80]. Actin is involved in the endocytic event and both CCPs and especially CCSs appear to act as platforms for cytoskeletal organization [77–80]. Disruption of the actin–clathrin cross-talk disorganizes the intermediate filament network [78,81]. CCPs are stiffer than the plasma membrane, which is partly due to the rigidification of the clathrin coat by accessory proteins such as the AP2 adaptor complex [82]. CCPs in some instances form from CCSs [3,4,75,76]. In a context- and size-dependent manner, CCPs and CCSs regulate cell adhesion, mechanotransduction, and endocytosis [3,4,75,76].

CCSs, in association with dynamin (DNM)2, a large GTPase that acts as a mechanochemical scaffolding molecule [3,4], sense and transduce mechanical stimuli at the plasma membrane and regulate YAP/TAZ activity [79]. Upon mechanical stimulation, YAP appears to associate with branching actin filaments and accumulate around CCSs and CCPs. Importantly, YAP/TAZ interacts with the central CCP component DNM2 [79] (Figure 2A,B). The spatiotemporal accumulation of YAP/TAZ at actin filaments surrounding clathrin-coated structures is an additional regulatory mechanotransductive complex. However, details on how YAP/TAZ are recruited to these plasma membrane structures remain to be elucidated.

αVβ5 integrins are central to CCS stability. The interaction of αVβ5 with the ligand vitronectin and their subsequent clustering is mediated via clathrin adaptor proteins, which is indispensable for assembly of most CCSs [83]. Knockdown of αVβ5 leads to loss of large and static CCSs, while overexpression results in more stable CCSs [84]. αVβ5 integrins are especially important as mechanotransducers on rigid surfaces. Inhibition of αVβ5 reduces YAP/TAZ activity, highlighting the importance of feedback regulation between integrins, clathrin, and the Hippo pathway (Figure 2C) [84,85].

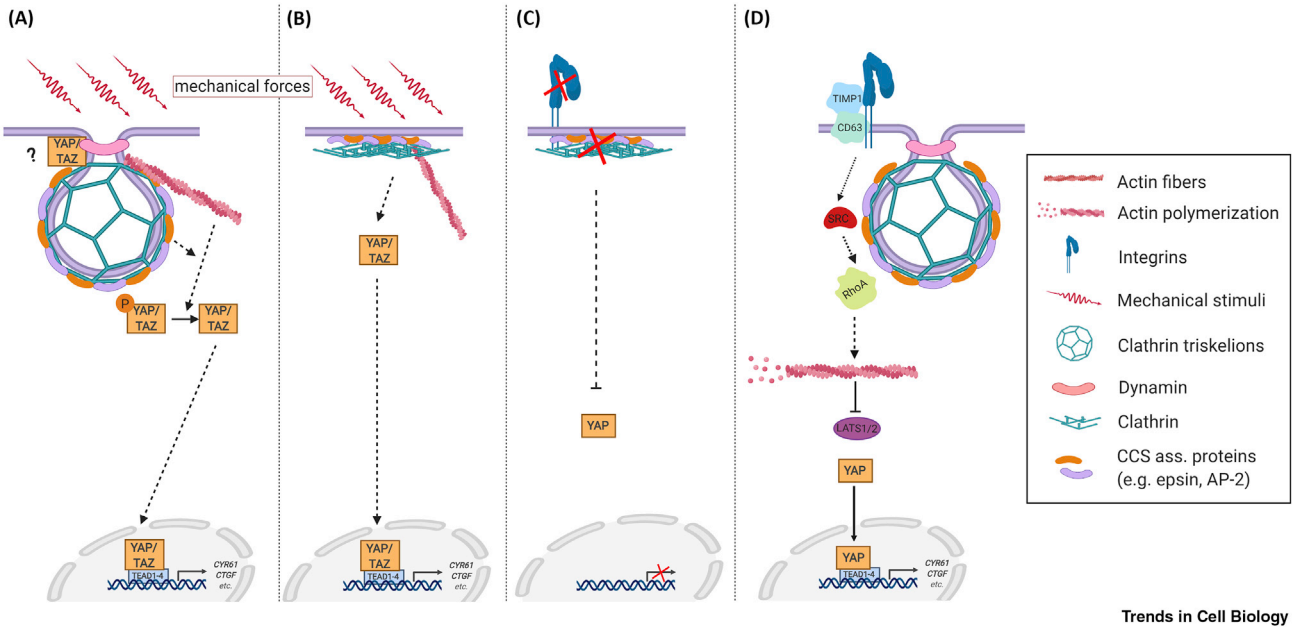

Figure 2. Hippo Pathway and Clathrin-Coated Pits and Plaques.
Clathrin-coated structures at the plasma membrane are endocytic active structures, but also function as adhesion complexes. Most interactions between YAP/TAZ and clathrin-coated pits (A and D) as well as plaques (B and C) lead to the activation of YAP (and TAZ), in particular, as a response to mechanical stress (A and B). (A) Mechanical stimuli induce association of YAP/TAZ with dynamin 2 and YAP/TAZ activity is increased by actin filaments accumulated around clathrin-coated structures (CCSs). (B) Mechanical forces are transduced at clathrin-coated pits (CCPs) via associated actin filaments and induce YAP/TAZ activity. (C) Inhibition of αVβ5 integrins reduces YAP/TAZ activity. (D) Interaction of tissue inhibitor of metalloproteinase 1 (TIMP1) with CD63 and integrin β1 activates SRC and RhoA-mediated actin assembly, leading to LATS1/2 inhibition and YAP/TAZ activation.

Moreover, in the absence of CCSs, αVβ5 integrins shuttle to FAs, functionally linking clathrin-coated structures with FAs [84–87]. Clathrin light chain (CLC)a is required for FAK localization at the adherent surface during cell spreading, as well as integrin-dependent FAK and SRC activation. CLCa deficiency inhibits FA maturation, emphasizing the importance of clathrin for plasma membrane mechanotransductive complexes [88]. The direct interplay between clathrin and FAs in the context of YAP/TAZ is yet to be explored.

Membrane-type 1 matrix metalloproteinases (MT1-MMPs) are endocytosed predominately in a clathrin-dependent manner [89]. Deficiency of MT1-MMP reduces ECM fibronectin proteolysis and integrin α5β1 endocytosis. This causes an imbalance between ECM turnover and overall increased ECM degradation [90,91]. Consequently, clathrin might influence ECM properties by mediating MT1-MMP turnover, which in turn influences YAP/TAZ activity. Furthermore, the tissue inhibitor of metalloproteinase (TIMP)1 complexes with MT1-MMP and blunts its enzymatic function [92]. TIMP1 expression is elevated in a range of cancers, where TIMP1 complexes with CD63 and integrin β1, which activates SRC and promotes RhoA-mediated actin assembly. As a consequence, LATS1/2 are inhibited, leading to activation of YAP/TAZ, favoring cell proliferation (Figure 2D) [93].

The cellular internalization rate of receptors commonly taken up via clathrin, such as VE-cadherin [94,95], shown in experiments with pulse-labeled VE-cadherin molecules at cell junctions is decreased upon YAP/TAZ depletion [94]. YAP/TAZ therefore appear to regulate the turnover of CCPs and CCSs. Further detailed analysis is needed to establish exactly how the endocytic machinery is regulated by YAP/TAZ. However, YAP/TAZ drive cytoskeletal dynamics [37,57–59] and clathrin-dependent endocytosis likewise relies on actin forces [4,80], and thus the cytoskeleton might be central in this regulation. How widespread this process is, and potential further cellular feedback still needs to be established. CCSs and CCPs engage in a range of cell-type-dependent and dynamic mechanosensory and endosomal functions, and consequently, the links to the Hippo pathway and YAP/TAZ activity are likewise expected to be versatile.

The Hippo Pathway, YAP/TAZ, and Caveolae

Caveolae are 50–80-nm invaginations of the plasma membrane and are composed of specialized lipids and caveolin (CAV)1–3, cavin 1–4, EHD, and pacsin proteins [2,5,96]. Caveolae-deficient patients and animal models have lipid and muscular dystrophies [97–102]. A principal function of caveolae is mechanosensing and -protection [96,99,103–107]. Endothelial cells, adipocytes, and myocytes need constantly to adapt to changes in mechanical stresses, and consequently it might not be coincidental that they harbor a large number of caveolae [108]. Under increased mechanical forces in stretched cells and upon osmotic shock, caveolae appear to flatten and thereby regulate cellular surface to volume ratio, which, at least in cell types with abundant caveolae, are thought to provide a plasma membrane buffering capacity (Box 3). In the process of flattening the membrane-associated part of the caveolar protein complex (e.g., cavins and EHDs) dissociates from the membrane [99,105,109]. Although on the surface there appear to be overlapping functions between caveolae and the Hippo pathway, a direct interdependence of central caveolae elements and the Hippo pathway has only recently been identified (Figure 3) [110,111].

YAP/TAZ are, via TEAD activation, essential for caveolae expression in both mammalian cell culture and in zebrafish [111]. In YAP/TAZ-deficient cells the expression levels of *CAVIN1* and *CAV1* are decreased by >85% [111]. This dramatic effect is caused by cell intrinsic activity and mediated via YAP/TAZ activation of TEAD. YAP/TAZ knockout therefore causes a remarkable loss of caveolae and YAP/TAZ-deficient cells therefore lose an entire cellular organelle [111]. Shear stress induces YAP/TAZ activity [112,113]; a process that is partly caveolae dependent [111] and transduced via the Hippo pathway, which regulates the transcription of ECM components [59,61,111]. Caveolae prevent rupture of endothelial plasma membranes under physiological hemodynamic force [103,109]. Healthy shear stress sensing and cellular response are crucial, as they shape the vascular system during development [114]. Dysfunctioning endothelial shear stress sensing and transduction in adult life is a major cause of atherosclerosis and vascular malformations [114]. It is possible that dysfunctioning

Box 3. Caveolae

Caveolae, Latin for little caves were discovered in the early 1950s by George Palade and Eichi Yamada [166,167]. Caveolae are 50–80-nm bulb-shaped actin-linked [168–170] plasma membrane invaginations present in the majority of cell types and most abundant in endothelial cells, myocytes, and adipocytes [2,5]. Structurally, caveolae are composed of membrane-embedded caveolins, the peripheral membrane cavins, as well as the associated elements EHDs, pacsins, and a specialized plasma membrane lipid composition [2,5,96]. The two essential proteins for caveolae assembly and stability are CAV1 (in nonmuscle cells) and cavin1 [2,5,96]. The current detailed caveolar location of EHDs (predominantly EHD2), cavins, and caveolins is well understood, whereas the location of the pacsin protein is less well characterized. Caveolae are structurally diverse, which is dictated, at least partly, by different ratios of distinct cavin complexes [2,108,171–173] (Figure III). Although in the past, caveolae were overwhelmingly categorized as clathrin-independent endocytic structures, this absolute view of caveolae has now changed, mainly due to the overall limited direct evidence of the endocytic event, and especially the lack of any caveolae specific cargoes, in *in vitro* cell cultures [2,17,174], to now include a broader role in mechanotransduction and -protection, regulation of membrane lipid composition, and cell signaling [5,96].

Upon increased cellular stretch, such as occurring under osmotic swelling and mechanical stretches, caveolae appear to flatten [104,105,175,176] (Figure III). While the plasma-membrane-embedded caveolins are retained in the plasma membrane, the cavin complexes and additional associated functional caveolae proteins are released into the cytosol. In the cardiovascular system, hemodynamic forces act tangentially on endothelial cells [152]. In endothelial cells, caveolae influence blood vessel remodeling and harbor mechanoprotective properties [103,109]. Increased blood flow induces flattening of caveolae and the apparent release of caveolae components, such as cavins and EHDs, into the cytoplasm [109]. In particular, proteins of the EHD family, located at the neck of the caveolae bulb, are important mechanotransductive caveolae components [173,177–182]. Released EHD2 is SUMOylated and translocates to the nucleus, where it binds to and in a context-dependent manner represses or activates the transcription factors Krüppel-like factor (KLF)7 [182,183] and modulator of KLF7 activity (MoKA) [182]. This transcriptional complex activates TNF-α, K-Ras as well as some caveolar genes, which appears to be an EHD2-mediated feedback mechanism to induce caveolae reconstitution [182,183].

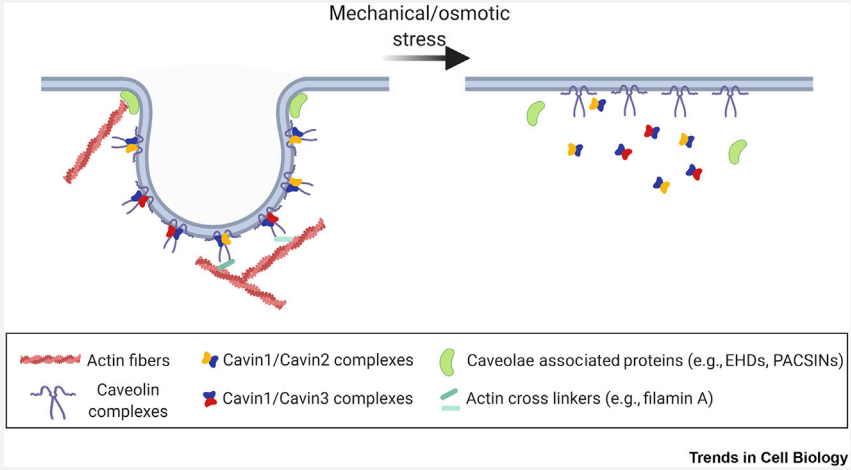

Figure III. Caveolae Organization and Dynamics.

interplay between the Hippo pathway and caveolae underlies these disease states. Although the exact mechanism by which caveolae regulate the Hippo pathway is incomplete, a few plausible mechanisms exist. In myoblasts stretch-induced caveolae disassembly leads to SRC activation [106], but whether this SRC activation mediates YAP activation via LATS inhibition, or by directly activating phosphorylation of YAP directly (Box 1) is still to be explored. The plasmalemmal anionic lipid, phosphatidylserine (PS), is an important mediator of cellular signaling [115]. PS is required for caveolae formation [116]. Multiple caveolae components bind to PS and especially the cytosolic recruitment of the

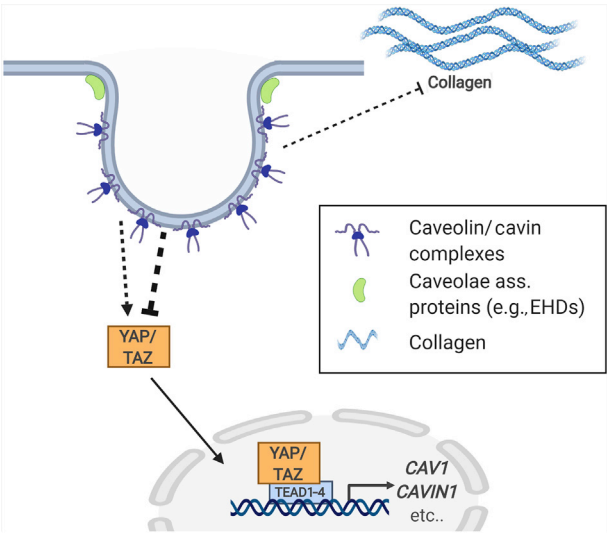

Figure 3. Hippo Pathway and Caveolae.
Caveolae are composed of caveolin complexes, distinct cavin complexes, EHD, and pacsin proteins. Caveolae regulate YAP/TAZ in a context- and cell-type-dependent manner. YAP/TAZ–TEAD transcriptionally induce the essential caveolar genes encoding *CAV1* and *CAVIN1*, which provide context dependent cellular feedback. As examined in both CAV1 and CAVIN1 deficient *in vitro* and *in vivo* models, caveolae are inhibitors of the expression of a range of extracellular matrix proteins, including various collagens.

integral caveolar proteins, the cavins, to the plasma membrane proteins appear to depend on the increased avidity upon PS clustering [2,115]. CAV1 deficiency results in altered cellular lipid composition, and plasma membrane PS distribution [117]. PS in recycling endosomes activates YAP [118]. The redistributed PS in CAV1-deficient cells might contribute to the altered YAP activity in CAV1-deficient cells.

Hypo-osmotic stress flattens caveolae (Box 3) and also regulates the Hippo pathway. Osmotic stress stimulates transient YAP nuclear localization by Nemo-like kinase (NLK)-mediated Ser128 phosphorylation of YAP. YAP phosphorylation on Ser127 and Ser128 appears mutually exclusive [119,120]. This antagonizes and disrupts the inhibitory complex formation between YAP and 14-3-3, and increases YAP activity even when YAP is phosphorylated at the LATS1/2-mediated Ser127 [119,120]. However, how and if caveolae and the Hippo pathway are interlinked in the cellular response to osmotic stress is currently not established.

In addition, mechanoprotective properties of caveolae also take place in hemidesmosomes (HDs). HDs are epithelial-specific plasma membrane complexes anchoring the cell's keratin network to the ECM and protecting the cell from mechanical stress [121]. The HD-specific α6β4 integrin [121] is cotransported with CAV1 during remodeling processes [122]. Upon cell stretching and hypo-osmotic shock, α6β4 is released from HDs. The subsequent trafficking of HD integrins is (indirectly) dependent on caveolae [122], indicating that caveolae are required for HD generation and turnover. Moreover, α6β4 activates YAP via laminin 322 [123], which additionally might link caveolae and the Hippo pathway.

Caveolae modulate ECM homeostasis and composition. In mammary glands CAV1 knockout causes increased expression of ECM components, such as fibronectin, tenascin C, and collagens, which increase ECM stiffness [124]. Cavin1 deficiency in adipocytes likewise increases expression of fibronectin and collagens [125]. Furthermore, CRISPR genome edited CAV1-deficient NIH3T3 cells (mouse

fibroblasts) produce, due to increased gene expression, elevated levels of ECM components, such as elastins and collagens (Figure 3) [126]. Increased levels of ECM due to decreased caveolae abundance might underlie the lung fibrosis observed in CAV1-deficient animals and in clinical manifestations such as idiopathic pulmonary fibrosis [127,128]. Codependence of protein stability between CAV1 and CAVIN1 might therefore also explain the increase in collagen and fibronectin expression in CAV1- and CAVIN1-deficient cell lines and animal models [97,98,108,124–126]. Caveolae-dependent β1 integrin endocytosis is furthermore a mediator of fibronectin matrix turnover. CAV1-deficient cells have decreased levels of β1 integrin and fibronectin internalization and increased plasma membrane β1 integrin [129]. β1 integrin is an inhibitor of LATS1/2 and consequently activates YAP, but β1 integrin also appears to activate YAP independently of LATS [36,130]. The ECM composition dictates Hippo pathway activity and increased ECM rigidity activates YAP/TAZ [11]. It is therefore plausible that the increased activation of YAP/TAZ in some CAV1 knockout cells might be caused by increased synthesis of ECM. However, in mouse embryonic fibroblasts, CAV1 activates YAP in response to substrate stiffness in an actin-cytoskeleton-dependent manner, which might be an alternate and context dependent way for the cell to adapt to changes in ECM stiffness [110].

Mechanical cues as well as YAP/TAZ activity regulate senescence [131]. YAP/TAZ bypass senescence induced by mechanical cues by altering nucleotide metabolism [131]. Regulation of senescence is furthermore regulated by CAV1 and K-Ras. CAV1 deficiency reduces oncogenic K-Ras-induced premature senescence. K-Ras also induces the inhibitory interaction of CAV1 with the central detoxifier MTH1 limiting senescence [132]. Furthermore, in pancreatic cancer, K-Ras regulates YAP activity and YAP is a well-characterized negative regulator of senescence [133,134]. Consequently, caveolae might also be linked to YAP activity via K-Ras and regulation of senescence; however, a potential causative interaction is still missing.

In conclusion, both caveolae and the Hippo pathway are key regulators of central cellular processes [5,12,110,111]. The direct regulation of essential caveolar genes by the Hippo pathway [111] highlights the importance of this robust cellular feedback [110,111].

The Hippo Pathway, YAP/TAZ, and Primary Cilia

Primary cilia are nonmotile plasma membrane evaginations and play central cellular roles in sensing both mechanical (cilium bending) and chemical stimuli [7,135].

YAP/TAZ and the primary cilia are reciprocal negative regulators. Cytoplasmic retention or YAP/TAZ deficiency correlates with cell rounding, smaller cell size, and increased cilia formation [33,67,136–139]. The kinases LIMK2 and TESK1 are actin-remodeling factors that suppress ciliogenesis by inhibiting ciliary vesicle trafficking at least partly via YAP/TAZ activation (Figure 4A) [139]. MST1 is activated during ciliogenesis and localizes to the basal body of cilia, where MST1/2 facilitate development of mature primary cilia by promoting localization of multiple ciliary cargoes (e.g,. RAB8A, Smo, and RPGR) [140]. Aurora kinase (AURK)A localizes to the basal body of the cilium and induces ciliary resorption in response to growth factor stimulation [141]. MST1/2 mediate direct phosphorylation of AURKA, which interferes with formation of the AURKA/HDAC6 cilia-disassembly complex. MST1/2 thereby inhibit primary cilia disassembly [140] (Figure 4B). Similarly, expression of the primary cilia disassembly factors AURKA and PLK1 are increased upon YAP activation (Figure 4C) [140]. Complementary experiments using cytochalasin D, an actin destabilizer that causes cytoplasmic YAP/TAZ, also induces ciliogenesis and elongated cilia (Figure 4D) [139]. The cilia-associated proteins nephrocystin (NPHP)4 and NPHP9 inhibit the Hippo kinase cascade [137,138]. NPHP4 binds to and inhibits LATS1, whereby NPHP4 facilitates YAP/TAZ-mediated cell proliferation (Figure 4E) [137]. Inhibition of YAP/TAZ expression and their cytoplasmic retention have been linked to cilia disassembly, indicating the prominent role of YAP/TAZ activity in suppressing ciliogenesis.

Primary cilia are cellular sensory modalities, and links between the Hippo pathway and this cellular antenna need further exploration. YAP/TAZ drive the expression of a range of prominent signaling

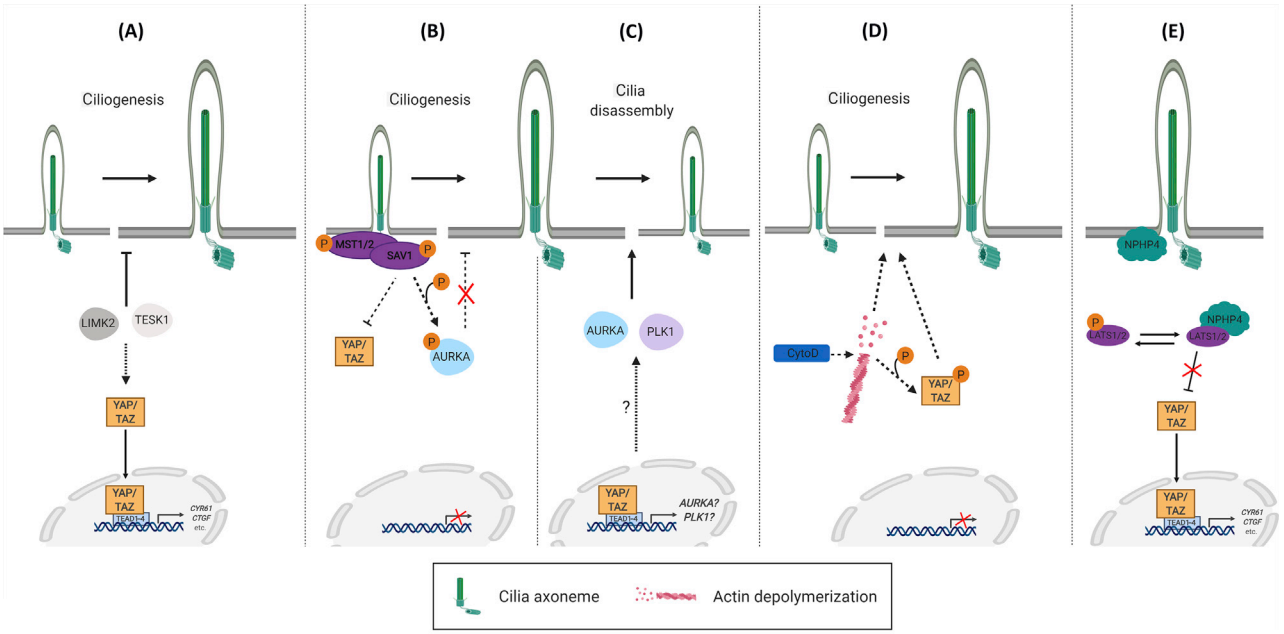

Figure 4. Hippo Pathway and Primary Cilia.

Primary cilia are microtubule-based organelles that coordinate signal transduction. Several components involved in cilia formation and cilia disassembly also regulate YAP/TAZ activity. (A) The kinases LIMK2 and TESK1 inhibit ciliogenesis (at least partly) via YAP/TAZ activation. (B) MST1/2 facilitate development and stability of mature primary cilia by inhibition of YAP/TAZ activity and inhibitory phosphorylation of Aurora kinase A (AURKA). (C) YAP activation increases the expression of the primary cilia disassembly factors AURKA and PLK1. (D) The actin destabilizer cytochalasin D (CytoD) causes YAP/TAZ inactivation and induces ciliogenesis and elongated cilia. (E) The cilia-associated protein nephrocystin-4 (NPHP4) binds to LATS1 and inhibits LATS-mediated phosphorylation of YAP/TAZ. Consequently YAP/TAZ is activated.

ligands and receptors [10], such as those involved in Wnt, BMP, Notch, growth factors, and transforming growth factor (TGF)-β signaling, which also play key roles in primary cilia biogenesis and function [7,135,142]. Importantly, YAP/TAZ activity might be indirectly regulated by the interdependent interactions between ciliogenesis and caveolae. Pacsins and EHDs stabilize and regulate the dynamics of caveolae [2,5,96,143] as well as facilitate tubulation in ciliogenesis [144,145]. In immature cilia, pacsins function at ciliary vesicles promoting the transition of the mother centriole to the basal body, while in mature cilia both pacsins and EHDs are at the ciliary pocket membrane and leave the cilium via membrane tubules [145]. A CAV1 isoform (CAV1α) inhibits primary cilia length elongation via ROCK-mediated regulation of RhoA [146], which might be caused by a CAV1-dependent failure in resorption of the mature primary cilia [147]. It is therefore important to acknowledge the challenges in determining what is correlative and what is causative, in order to decipher the full molecular functional interactions of these prominent cellular signaling nexuses with the Hippo pathway.

Concluding Remarks

The plasma membrane is a dynamic chemical and mechanical transducer. Adaptable sensing of extracellular signals is essential in order to ensure context-specific cell homeostasis and regulate differentiation. Tight and adherens junctions are established major intercellular regulators of the Hippo pathway [10,15,16]. In addition **desmosomes** [148], specialized adhesive protein complexes that also localize to intercellular junctions, likewise regulate YAP/TAZ [149,150]. Dysregulation of desmosomes causes context-dependent YAP regulation [149,150], and multiple desmosome components are likely YAP/TAZ–TEAD target genes, as informed by their presence in large scale chromatin immunoprecipitation (ChIP)-seq analysis of Tead4 target genes [151], providing further cellular feedback.

Key Figure

Hippo Pathway and Plasma Membrane Interactions

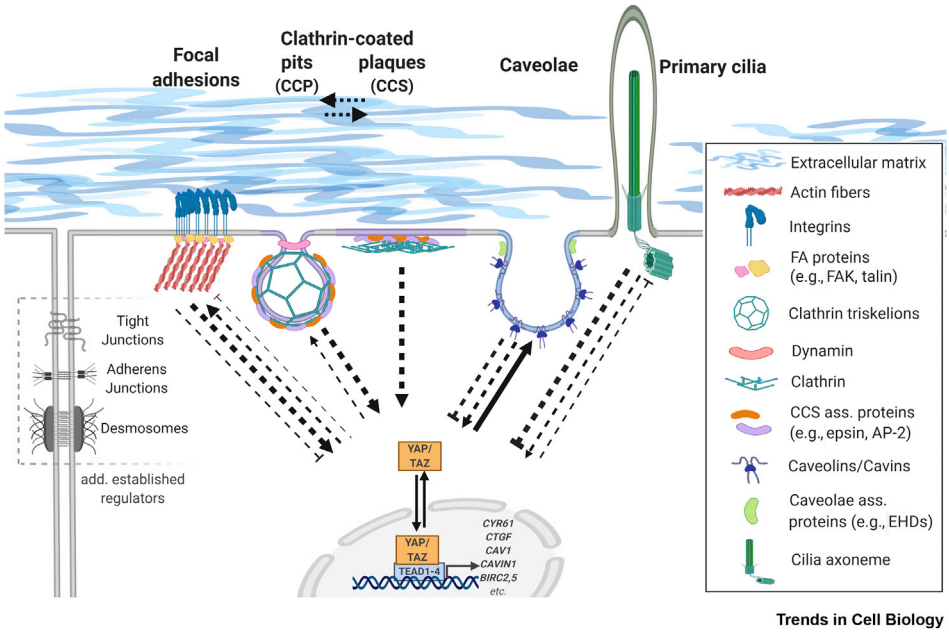

Figure 5. Focal adhesions (FAs), clathrin-coated pits (CCPs) and plaques (CCSs), as well as caveolae and primary cilia have strong reciprocal interactions with the Hippo pathway, resulting in a context dependent activation or inhibition of YAP/TAZ activity directed from the plasma membrane. The weight and direction of the arrows indicate the collective reported scale of the effects. Also depicted are cellular junctions (tight junctions, adherens junctions, and desmosomes) as additional established regulators of the Hippo pathway. Abbreviations: add., additional; ass., associated.

Additional, highly specific plasma membrane elements, such as FAs, CCPs and plaques, caveolae, and primary cilia are key to enable a defined localized response to a large variety of signals [3–7]. Those plasma membrane structures dynamically sense and transmit chemical and mechanical stimuli to the Hippo pathway (Figure 5, Key Figure) [2–8,11]. Most cell types, in particular endothelial cells, myocytes, and bone cells, constantly experience mechanical forces through shear stress and tension [6,9,11,152]. The Hippo pathway is a nexus and integrator of cellular responses to tension, stretching, and changes of ECM properties [11,33] and contains multiple levels of robust cellular feedback loops, which ensures cellular homeostasis [37,111,153]. YAP/TAZ–TEAD also directly regulate the expression of central components of additional cell signaling pathways, such as Wnt, TGF, Notch, and BMP [10], and several of these have direct roles in for instance the primary cilia [7,135,142]. Importantly, the central plasma membrane components discussed here also interact with each other in order to respond to complex stimuli. This widespread spatiotemporal interplay between prominent members of apparent distinct plasma membrane domains highlight the intricate complexity of the plasma membrane, and therefore, also ultimately their dynamic interactions with the Hippo pathway. Furthermore, due to the wide range of transcriptionally regulated YAP/TAZ target genes, and consequently, the central role of YAP/TAZ in most cellular processes, it remains challenging to study the effect of potential additional roles of upstream Hippo pathway kinases and scaffolding proteins in regulating these cellular plasma membrane domains separately and independently of YAP/TAZ

Outstanding Questions

How are the cellular Hippo pathway components spatiotemporally regulated?

Subcellular localization drives compartmentalization and protein function. Specific Hippo pathway components temporally localize to junctional complexes. However, precise and dynamic information on the subcellular localization of Hippo pathway components in general, especially in vertebrates, is currently not well established. It is anticipated that Hippo pathway components might also be subcellularly distributed to additional plasma membrane domains. Genome editing, live cell imaging, and super-resolution microscopy at endogenous levels appear to be fertile avenues to explore. Caution must be taken to ensure that genome-tagged versions of the Hippo pathway components conserve their cellular dynamics and functionality. Recently developed proximity-dependent chemical biology approaches, such as APEX, BioID, as well as bio-orthogonal noncanonical amino acid tagging (*BONCAT*) might prompt further insights into the interplay between these signaling modalities.

Does the Hippo pathway alter the plasma membrane glycocalyx and lipid composition?

Determining the impact that YAP/TAZ activity has on the cellular glycocalyx and plasma membrane lipid composition could reveal fundamental new insights.

How widespread is Hippo pathway 'moonlighting'?

It is becoming clear that Hippo pathway core kinase components integrate additional seemingly independent substrates into context-specific signaling. The field might need to brace itself for surprising discoveries, where Hippo pathway components moonlight in distinct functions away from direct regulation of the core Hippo pathway. However, it remains

regulation. It should be acknowledged that obtaining mechanistic insights to decipher the importance of one type of regulation and to distinguish that from other inputs is conceptually challenging.

The chemical and mechanical clues cells respond to are diverse. Interconnecting plasma membrane structures play central roles in sensing and relaying extracellular signals to the Hippo pathway. This compartmentalization allows for a dynamic and context-specific response. However, our current understanding of how this delicate and dynamic interplay is regulated is far from complete (see Outstanding Questions). Future discoveries will provide mechanistic insights into cellular processes relevant in development, regenerative medicine, and disease.

challenging to study these moonlighting functions, as the parallel transcriptional regulation via YAP/TAZ drives a multitude of cellular processes.

Does the Hippo pathway regulate endocytosis?

Endocytosis is a major cellular regulating hub. The various types of endocytosis help regulate the plasma membrane abundance of receptors, and function to internalize ligands and nutrients. Many toxins, bacteria, and viruses hijack and usurp the cellular entry machinery. One could hypothesize that pathogens might have evolved to also take advantage of the pro-proliferative and antiapoptotic gene sets regulated by YAP/TAZ.

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References

1. Nicolson, G.L. (2014) The fluid-mosaic model of membrane structure: still relevant to understanding the structure, function and dynamics of biological membranes after more than 40 years. *Biochim. Biophys. Acta* 1838, 1451–1466
2. Hansen, C.G. and Nichols, B.J. (2010) Exploring the caves: cavins, caveolins and caveolae. *Trends Cell Biol.* 20, 177–186
3. Lampe, M. *et al.* (2016) Clathrin coated pits, plaques and adhesion. *J. Struct. Biol.* 196, 48–56
4. McMahon, H.T. and Boucrot, E. (2011) Molecular mechanism and physiological functions of clathrin-mediated endocytosis. *Nat. Rev. Mol. Cell Biol.* 12, 517–533
5. Parton, R.G. (2018) Caveolae: structure, function, and relationship to disease. *Annu. Rev. Cell Dev. Biol.* 34, 111–136
6. Burridge, K. (2017) Focal adhesions: a personal perspective on a half century of progress. *FEBS J.* 284, 3355–3361
7. Anvarian, Z. *et al.* (2019) Cellular signalling by primary cilia in development, organ function and disease. *Nat. Rev. Nephrol.* 15, 199–219
8. Pinheiro, D. and Bellaiche, Y. (2018) Mechanical force-driven adherens junction remodeling and epithelial dynamics. *Dev. Cell* 47, 3–19
9. Vogel, V. (2018) Unraveling the mechanobiology of extracellular matrix. *Annu. Rev. Physiol.* 80, 353–387
10. Hansen, C.G. *et al.* (2015) YAP and TAZ: a nexus for Hippo signaling and beyond. *Trends Cell Biol.* 25, 499–513
11. Panciera, T. *et al.* (2017) Mechanobiology of YAP and TAZ in physiology and disease. *Nat. Rev. Mol. Cell Biol.* 18, 758–770
12. Moroishi, T. *et al.* (2015) The emerging roles of YAP and TAZ in cancer. *Nat. Rev. Cancer* 15, 73–79
13. Moya, I.M. and Halder, G. (2019) Hippo-YAP/TAZ signalling in organ regeneration and regenerative medicine. *Nat. Rev. Mol. Cell Biol.* 20, 211–226
14. Luo, J. and Yu, F.X. (2019) GPCR-Hippo signaling in cancer. *Cells* Published online May 8, 2019. https://doi.org/10.3390/cells8050426
15. Karaman, R. and Halder, G. (2018) Cell junctions in Hippo signaling. *Cold Spring Harb. Perspect. Biol.* Published online May 1, 2018. https://doi.org/10.1101/cshperspect.a028753
16. Fulford, A. *et al.* (2018) Upstairs, downstairs: spatial regulation of Hippo signalling. *Curr. Opin. Cell Biol.* 51, 22–32
17. Hansen, C.G. and Nichols, B.J. (2009) Molecular mechanisms of clathrin-independent endocytosis. *J. Cell Sci.* 122, 1713–1721
18. Kim, N.G. *et al.* (2011) E-cadherin mediates contact inhibition of proliferation through Hippo signaling-pathway components. *Proc. Natl. Acad. Sci. U. S. A.* 108, 11930–11935
19. Yang, C.C. *et al.* (2015) Differential regulation of the Hippo pathway by adherens junctions and apical-basal cell polarity modules. *Proc. Natl. Acad. Sci. U. S. A.* 112, 1785–1790
20. Liu, X. *et al.* (2013) PTPN14 interacts with and negatively regulates the oncogenic function of YAP. *Oncogene* 32, 1266–1273
21. Wang, W. *et al.* (2012) PTPN14 is required for the density-dependent control of YAP1. *Genes Dev.* 26, 1959–1971
22. Varelas, X. *et al.* (2010) The Crumbs complex couples cell density sensing to Hippo-dependent control of the TGF-beta-SMAD pathway. *Dev. Cell* 19, 831–844
23. Zhao, B. *et al.* (2011) Angiomotin is a novel Hippo pathway component that inhibits YAP oncoprotein. *Genes Dev.* 25, 51–63
24. Mana-Capelli, S. and McCollum, D. (2018) Angiomotins stimulate LATS kinase autophosphorylation and act as scaffolds that promote Hippo signaling. *J. Biol. Chem.* 293, 18230–18241
25. Wang, W. *et al.* (2011) Angiomotin-like proteins associate with and negatively regulate YAP1. *J. Biol. Chem.* 286, 4364–4370
26. Moleirinho, S. *et al.* (2017) Regulation of localization and function of the transcriptional co-activator YAP by angiomotin. *eLife*. Published online May 3, 2017. https://doi.org/10.7554/eLife.23966
27. Yin, F. *et al.* (2013) Spatial organization of Hippo signaling at the plasma membrane mediated by the tumor suppressor Merlin/NF2. *Cell* 154, 1342–1355
28. Li, Y. *et al.* (2015) Angiomotin binding-induced activation of Merlin/NF2 in the Hippo pathway. *Cell Res.* 25, 801–817
29. Mao, X. *et al.* (2017) CRB3 regulates contact inhibition by activating the Hippo pathway in mammary epithelial cells. *Cell Death Dis.* 8, e2546
30. Sun, Z. *et al.* (2016) Integrin-mediated mechanotransduction. *J. Cell Biol.* 215, 445–456

31. Kechagia, J.Z. *et al.* (2019) Integrins as biomechanical sensors of the microenvironment. *Nat. Rev. Mol. Cell Biol.* 20, 457–473
32. Kim, N.G. and Gumbiner, B.M. (2015) Adhesion to fibronectin regulates Hippo signaling via the FAK-Src-PI3K pathway. *J. Cell Biol.* 210, 503–515
33. Dupont, S. *et al.* (2011) Role of YAP/TAZ in mechanotransduction. *Nature* 474, 179–183
34. Zhao, B. *et al.* (2012) Cell detachment activates the Hippo pathway via cytoskeleton reorganization to induce anoikis. *Genes Dev.* 26, 54–68
35. Lachowski, D. *et al.* (2018) FAK controls the mechanical activation of YAP, a transcriptional regulator required for durotaxis. *FASEB J.* 32, 1099–1107
36. Elbediwy, A. *et al.* (2016) Integrin signalling regulates YAP and TAZ to control skin homeostasis. *Development* 143, 1674–1687
37. Mason, D.E. *et al.* (2019) YAP and TAZ limit cytoskeletal and focal adhesion maturation to enable persistent cell motility. *J. Cell Biol.* 218, 1369–1389
38. Kissil, J.L. *et al.* (2002) Merlin phosphorylation by p21-activated kinase 2 and effects of phosphorylation on merlin localization. *J. Biol. Chem.* 277, 10394–10399
39. Xia, H. *et al.* (2004) Focal adhesion kinase is upstream of phosphatidylinositol 3-kinase/Akt in regulating fibroblast survival in response to contraction of type I collagen matrices via a beta 1 integrin viability signaling pathway. *J. Biol. Chem.* 279, 33024–33034
40. McLeod, S.J. *et al.* (2004) The Rap GTPases regulate integrin-mediated adhesion, cell spreading, actin polymerization, and Pyk2 tyrosine phosphorylation in B lymphocytes. *J. Biol. Chem.* 279, 12009–12019
41. Meng, Z. *et al.* (2018) RAP2 mediates mechanoresponses of the Hippo pathway. *Nature* 560, 655–660
42. Meng, Z. *et al.* (2015) MAP4K family kinases act in parallel to MST1/2 to activate LATS1/2 in the Hippo pathway. *Nat. Commun.* 6, 8357
43. Li, Q. *et al.* (2014) The conserved misshapen-warts-Yorkie pathway acts in enteroblasts to regulate intestinal stem cells in *Drosophila*. *Dev. Cell* 31, 291–304
44. Yu, F.X. *et al.* (2012) Regulation of the Hippo-YAP pathway by G-protein-coupled receptor signaling. *Cell* 150, 780–791
45. Matthews, B.D. *et al.* (2006) Cellular adaptation to mechanical stress: role of integrins, Rho, cytoskeletal tension and mechanosensitive ion channels. *J. Cell Sci.* 119, 508–518
46. Arthur, W.T. and Burridge, K. (2001) RhoA inactivation by p190RhoGAP regulates cell spreading and migration by promoting membrane protrusion and polarity. *Mol. Biol. Cell* 12, 2711–2720
47. Biname, F. *et al.* (2016) Cancer-associated mutations in the protrusion-targeting region of p190RhoGAP impact tumor cell migration. *J. Cell Biol.* 214, 859–873
48. Frank, S.R. *et al.* (2018) p190 RhoGAP promotes contact inhibition in epithelial cells by repressing YAP activity. *J. Cell Biol.* 217, 3183–3201
49. Zhao, B. *et al.* (2007) Inactivation of YAP oncoprotein by the Hippo pathway is involved in cell contact inhibition and tissue growth control. *Genes Dev.* 21, 2747–2761
50. Elbediwy, A. *et al.* (2018) Enigma proteins regulate YAP mechanotransduction. *J. Cell Sci.* Published online November 22, 2018. https://doi.org/10.1242/jcs.221788
51. Crisp, M. *et al.* (2006) Coupling of the nucleus and cytoplasm: role of the LINC complex. *J. Cell Biol.* 172, 41–53
52. Makhija, E. *et al.* (2016) Nuclear deformability and telomere dynamics are regulated by cell geometric constraints. *Proc. Natl. Acad. Sci. U. S. A.* 113, E32–40.
53. Alisafaei, F. *et al.* (2019) Regulation of nuclear architecture, mechanics, and nucleocytoplasmic shuttling of epigenetic factors by cell geometric constraints. *Proc. Natl. Acad. Sci. U. S. A.* 116, 13200–13209
54. Elosegui-Artola, A. *et al.* (2017) Force triggers YAP nuclear entry by regulating transport across nuclear pores. *Cell* 171, 1397–1410.e1314.
55. Guilluy, C. *et al.* (2014) Isolated nuclei adapt to force and reveal a mechanotransduction pathway in the nucleus. *Nat. Cell Biol.* 16, 376–381
56. Nardone, G. *et al.* (2017) YAP regulates cell mechanics by controlling focal adhesion assembly. *Nat. Commun.* 8, 15321
57. Porazinski, S. *et al.* (2015) YAP is essential for tissue tension to ensure vertebrate 3D body shape. *Nature* 521, 217–221
58. Qiao, Y. *et al.* (2017) YAP regulates actin dynamics through ARHGAP29 and promotes metastasis. *Cell Rep.* 19, 1495–1502
59. Zhao, B. *et al.* (2008) TEAD mediates YAP-dependent gene induction and growth control. *Genes Dev.* 22, 1962–1971
60. Shen, J. *et al.* (2018) Hippo component YAP promotes focal adhesion and tumour aggressiveness via transcriptionally activating THBS1/FAK signalling in breast cancer. *J. Exp. Clin. Cancer Res.* 37, 175
61. Calvo, F. *et al.* (2013) Mechanotransduction and YAP-dependent matrix remodelling is required for the generation and maintenance of cancer-associated fibroblasts. *Nat. Cell Biol.* 15, 637–646
62. Lo, C.M. *et al.* (2000) Cell movement is guided by the rigidity of the substrate. *Biophys. J.* 79, 144–152
63. Plotnikov, S.V. *et al.* (2012) Force fluctuations within focal adhesions mediate ECM-rigidity sensing to guide directed cell migration. *Cell* 151, 1513–1527
64. Puleo, J.I. *et al.* (2019) Mechanosensing during directed cell migration requires dynamic actin polymerization at focal adhesions. *J. Cell Biol.* Published online October 8, 2019. https://doi.org/10.1083/jcb.201902101
65. Xu, J. *et al.* (2018) Yorkie Functions at the cell cortex to promote myosin activation in a non-transcriptional manner. *Dev. Cell* 46, 271–284 e275
66. Hu, J.K. *et al.* (2017) An FAK-YAP-mTOR signaling axis regulates stem cell-based tissue renewal in mice. *Cell Stem Cell* 21, 91–106 e106
67. Hansen, C.G. *et al.* (2015) The Hippo pathway effectors YAP and TAZ promote cell growth by modulating amino acid signaling to mTORC1. *Cell Res.* 25, 1299–1313
68. Tumaneng, K. *et al.* (2012) YAP mediates crosstalk between the Hippo and PI(3)K-TOR pathways by suppressing PTEN via miR-29. *Nat. Cell Biol.* 14, 1322–1329
69. Park, Y.Y. *et al.* (2016) Yes-associated protein 1 and transcriptional coactivator with PDZ-binding motif activate the mammalian target of rapamycin complex 1 pathway by regulating amino acid transporters in hepatocellular carcinoma. *Hepatology* 63, 159–172
70. Nicklin, P. *et al.* (2009) Bidirectional transport of amino acids regulates mTOR and autophagy. *Cell* 136, 521–534
71. Henderson, N.C. *et al.* (2004) CD98hc (SLC3A2) interaction with beta 1 integrins is required for transformation. *J. Biol. Chem.* 279, 54731–54741

72. Estrach, S. *et al.* (2014) CD98hc (SLC3A2) loss protects against ras-driven tumorigenesis by modulating integrin-mediated mechanotransduction. *Cancer Res.* 74, 6878–6889
73. Yan, R. *et al.* (2019) Structure of the human LAT1-4F2hc heteromeric amino acid transporter complex. *Nature* 568, 127–130
74. Sachs, N. *et al.* (2012) Loss of integrin alpha3 prevents skin tumor formation by promoting epidermal turnover and depletion of slow-cycling cells. *Proc. Natl. Acad. Sci. U. S. A.* 109, 21468–21473
75. Sochacki, K.A. and Taraska, J.W. (2019) From flat to curved clathrin: controlling a plastic ratchet. *Trends Cell Biol.* 29, 241–256
76. Yoshida, A. *et al.* (2018) Morphological changes of plasma membrane and protein assembly during clathrin-mediated endocytosis. *PLoS Biol.* 16, e2004786
77. Heuser, J. (1980) Three-dimensional visualization of coated vesicle formation in fibroblasts. *J. Cell Biol.* 84, 560–583
78. Vassilopoulos, S. *et al.* (2014) Actin scaffolding by clathrin heavy chain is required for skeletal muscle sarcomere organization. *J. Cell Biol.* 205, 377–393
79. Franck, A. *et al.* (2019) Mechanosensitive clathrin platforms anchor desmin intermediate filaments in skeletal muscle. *Biorxiv* Preprint Published online July 18, 2018. https://doi.org/10.1101/321885
80. Taylor, M.J. *et al.* (2011) A high precision survey of the molecular dynamics of mammalian clathrin-mediated endocytosis. *PLoS Biol.* 9, e1000604
81. Franck, A. *et al.* (2019) Clathrin plaques and associated actin anchor intermediate filaments in skeletal muscle. *Mol. Biol. Cell* 30, 579–590
82. Lherbette, M. *et al.* (2019) The AP2 adaptor enhances clathrin coat stiffness. *FEBS J.* Published online June 14, 2019. https://doi.org/10.1111/febs.14961
83. Zuidema, A. *et al.* (2018) Mechanisms of integrin alphaVbeta5 clustering in flat clathrin lattices. *J. Cell Sci.* Published online November 5, 2018. https://doi.org/10.1242/jcs.221317
84. Baschieri, F. *et al.* (2018) Frustrated endocytosis controls contractility-independent mechanotransduction at clathrin-coated structures. *Nat. Commun.* 9, 3825
85. Gvozdenovic, A. *et al.* (2016) Targeting αvβ3 and αvβ5 integrins inhibits pulmonary metastasis in an intratibial xenograft osteosarcoma mouse model. *Oncotarget* 7, 55141–55154
86. Ezratty, E.J. *et al.* (2009) Clathrin mediates integrin endocytosis for focal adhesion disassembly in migrating cells. *J. Cell Biol.* 187, 733–747
87. Yu, C.H. *et al.* (2015) Integrin-beta3 clusters recruit clathrin-mediated endocytic machinery in the absence of traction force. *Nat. Commun.* 6, 8672
88. Tsygankova, O.M. and Keen, J.H. (2019) A unique role for clathrin light chain A in cell spreading and migration. *J. Cell Sci.* Published online May 15, 2019. https://doi.org/10.1242/jcs.224030
89. Jiang, A. *et al.* (2001) Regulation of membrane-type matrix metalloproteinase 1 activity by dynamin-mediated endocytosis. *Proc. Natl. Acad. Sci. U. S. A.* 98, 13693–13698
90. Song, T. *et al.* (2015) TIMP-1 activated carcinoma-associated fibroblasts inhibit tumor apoptosis by activating SDF1/CXCR4 signaling in hepatocellular carcinoma. *Oncotarget* 6, 12061–12079
91. Shi, F. and Sottile, J. (2011) MT1-MMP regulates the turnover and endocytosis of extracellular matrix fibronectin. *J. Cell Sci.* 124, 4039–4050
92. Egea, V. *et al.* (2012) Tissue inhibitor of metalloproteinase-1 (TIMP-1) regulates mesenchymal stem cells through let-7f microRNA and Wnt/beta-catenin signaling. *Proc. Natl. Acad. Sci. U. S. A.* 109, E309–316
93. Ando, T. *et al.* (2018) Tissue inhibitor of metalloproteinase-1 promotes cell proliferation through YAP/TAZ activation in cancer. *Oncogene* 37, 263–270
94. Neto, F. *et al.* (2018) YAP and TAZ regulate adherens junction dynamics and endothelial cell distribution during vascular development. *eLife* Published online February 5, 2018. https://doi.org/10.7554/eLife.31037
95. Su, W. and Kowalczyk, A.P. (2017) The VE-cadherin cytoplasmic domain undergoes proteolytic processing during endocytosis. *Mol. Biol. Cell* 28, 76–84
96. Lamaze, C. *et al.* (2017) The caveolae dress code: structure and signaling. *Curr. Opin. Cell Biol.* 47, 117–125
97. Liu, L. *et al.* (2008) Deletion of Cavin/PTRF causes global loss of caveolae, dyslipidemia, and glucose intolerance. *Cell Metab.* 8, 310–317
98. Ding, S.Y. *et al.* (2017) Muscular dystrophy in PTFR/cavin-1 null mice. *JCI Insight* 2, e91023
99. Lo, H.P. *et al.* (2015) The caveolin-cavin system plays a conserved and critical role in mechanoprotection of skeletal muscle. *J. Cell Biol.* 210, 833–849
100. Hayashi, Y.K. *et al.* (2009) Human PTRF mutations cause secondary deficiency of caveolins resulting in muscular dystrophy with generalized lipodystrophy. *J. Clin. Invest.* 119, 2623–2633
101. Betz, R.C. *et al.* (2001) Mutations in CAV3 cause mechanical hyperirritability of skeletal muscle in rippling muscle disease. *Nat. Genet.* 28, 218–219
102. Cai, C. *et al.* (2009) Membrane repair defects in muscular dystrophy are linked to altered interaction between MG53, caveolin-3, and dysferlin. *J. Biol. Chem.* 284, 15894–15902
103. Yu, J. *et al.* (2006) Direct evidence for the role of caveolin-1 and caveolae in mechanotransduction and remodeling of blood vessels. *J. Clin. Invest.* 116, 1284–1291
104. Kozera, L. *et al.* (2009) Caveolae act as membrane reserves which limit mechanosensitive I(Cl,swell) channel activation during swelling in the rat ventricular myocyte. *PLoS One* 4, e8312
105. Sinha, B. *et al.* (2011) Cells respond to mechanical stress by rapid disassembly of caveolae. *Cell* 144, 402–413
106. Gervasio, O.L. *et al.* (2011) Caveolae respond to cell stretch and contribute to stretch-induced signaling. *J. Cell Sci.* 124, 3581–3590
107. Joshi, B. *et al.* (2012) Phosphocaveolin-1 is a mechanotransducer that induces caveola biogenesis via Egr1 transcriptional regulation. *J. Cell Biol.* 199, 425–435
108. Hansen, C.G. *et al.* (2013) Deletion of cavin genes reveals tissue-specific mechanisms for morphogenesis of endothelial caveolae. *Nat. Commun.* 4, 1831
109. Cheng, J.P. *et al.* (2015) Caveolae protect endothelial cells from membrane rupture during increased cardiac output. *J. Cell Biol.* 211, 53–61
110. Moreno-Vicente, R. *et al.* (2018) Caveolin-1 modulates mechanotransduction responses to substrate stiffness through actin-dependent control of YAP. *Cell Rep.* 25, 1622–1635 e1626
111. Rausch, V. *et al.* (2019) The Hippo pathway regulates caveolae expression and mediates flow response via caveolae. *Curr. Biol.* 29, 242–255 e246
112. Wang, K.C. *et al.* (2016) Flow-dependent YAP/TAZ activities regulate endothelial phenotypes and atherosclerosis. *Proc. Natl. Acad. Sci. U. S. A.* 113, 11525–11530

113. Lee, H.J. *et al.* (2017) Fluid shear stress activates YAP1 to promote cancer cell motility. *Nat. Commun.* 8, 14122
114. Baeyens, N. *et al.* (2016) Endothelial fluid shear stress sensing in vascular health and disease. *J. Clin. Invest.* 126, 821–828
115. Skotland, T. and Sandvig, K. (2019) The role of PS 18:0/18:1 in membrane function. *Nat. Commun.* 10, 2752
116. Hirama, T. *et al.* (2017) Phosphatidylserine dictates the assembly and dynamics of caveolae in the plasma membrane. *J. Biol. Chem.* 292, 14292–14307
117. Ariotti, N. *et al.* (2014) Caveolae regulate the nanoscale organization of the plasma membrane to remotely control Ras signaling. *J. Cell Biol.* 204, 777–792
118. Matsudaira, T. *et al.* (2017) Endosomal phosphatidylserine is critical for the YAP signalling pathway in proliferating cells. *Nat. Commun.* 8, 1246
119. Hong, A.W. *et al.* (2017) Osmotic stress-induced phosphorylation by NLK at Ser128 activates YAP. *EMBO Rep.* 18, 72–86
120. Moon, S. *et al.* (2017) Phosphorylation by NLK inhibits YAP-14-3-3-interactions and induces its nuclear localization. *EMBO Rep.* 18, 61–71
121. Manso, J.A. *et al.* (2019) Integrin alpha6beta4 recognition of a linear motif of bullous pemphigoid antigen BP230 controls its recruitment to hemidesmosomes. *Structure* 27, 952–964 e956
122. Osmani, N. *et al.* (2018) An Arf6- and caveolae-dependent pathway links hemidesmosome remodeling and mechanoresponse. *Mol. Biol. Cell* 29, 435–451
123. De Rosa, L. *et al.* (2019) Laminin 332-dependent YAP dysregulation depletes epidermal stem cells in junctional epidermolysis bullosa. *Cell Rep.* 27, 2036–2049.e2036.
124. Thompson, C. *et al.* (2017) Loss of caveolin-1 alters extracellular matrix protein expression and ductal architecture in murine mammary glands. *PLoS One* 12, e0172067
125. Ding, S.Y. *et al.* (2014) Pleiotropic effects of cavin-1 deficiency on lipid metabolism. *J. Biol. Chem.* 289, 8473–8483
126. Mendoza-Topaz, C. *et al.* (2018) Cells respond to deletion of CAV1 by increasing synthesis of extracellular matrix. *PLoS One* 13, e0205306
127. Wang, X.M. *et al.* (2006) Caveolin-1: a critical regulator of lung fibrosis in idiopathic pulmonary fibrosis. *J. Exp. Med.* 203, 2895–2906
128. Drab, M. *et al.* (2001) Loss of caveolae, vascular dysfunction, and pulmonary defects in caveolin-1 gene-disrupted mice. *Science* 293, 2449–2452
129. Shi, F. and Sottile, J. (2008) Caveolin-1-dependent beta1 integrin endocytosis is a critical regulator of fibronectin turnover. *J. Cell Sci.* 121, 2360–2371
130. Sabra, H. *et al.* (2017) beta1 integrin-dependent Rac/group I PAK signaling mediates YAP activation of Yes-associated protein 1 (YAP1) via NF2/merlin. *J. Biol. Chem.* 292, 19179–19197
131. Santinon, G. *et al.* (2018) dNTP metabolism links mechanical cues and YAP/TAZ to cell growth and oncogene-induced senescence. *EMBO J.* Published online June 1, 2018. https://doi.org/10.15252/embj.201797780
132. Volonte, D. *et al.* (2018) Caveolin-1 promotes the tumor suppressor properties of oncogene-induced cellular senescence. *J. Biol. Chem.* 293, 1794–1809
133. Xie, Q. *et al.* (2013) YAP/TEAD-mediated transcription controls cellular senescence. *Cancer Res.* 73, 3615–3624
134. Zhang, W. *et al.* (2014) Downstream of mutant KRAS, the transcription regulator YAP is essential for neoplastic progression to pancreatic ductal adenocarcinoma. *Sci. Signal.* 7, ra42
135. Malicki, J.J. and Johnson, C.A. (2017) The cilium: cellular antenna and central processing unit. *Trends Cell Biol.* 27, 126–140
136. Boehlke, C. *et al.* (2010) Primary cilia regulate mTORC1 activity and cell size through Lkb1. *Nat. Cell Biol.* 12, 1115–1122
137. Habbig, S. *et al.* (2011) NPHP4, a cilia-associated protein, negatively regulates the Hippo pathway. *J. Cell Biol.* 193, 633–642
138. Habbig, S. *et al.* (2012) The ciliopathy disease protein NPHP9 promotes nuclear delivery and activation of the oncogenic transcriptional regulator TAZ. *Hum. Mol. Genet.* 21, 5528–5538
139. Kim, J. *et al.* (2015) Actin remodelling factors control ciliogenesis by regulating YAP/TAZ activity and vesicle trafficking. *Nat. Commun.* 6, 6781
140. Kim, M. *et al.* (2014) The MST1/2-SAV1 complex of the Hippo pathway promotes ciliogenesis. *Nat. Commun.* 5, 5370
141. Pugacheva, E.N. *et al.* (2007) HEF1-dependent Aurora A activation induces disassembly of the primary cilium. *Cell* 129, 1351–1363
142. Satir, P. and Satir, B.H. (2019) The conserved ancestral signaling pathway from cilium to nucleus. *J. Cell Sci.* Published online August 2, 2019. https://doi.org/10.1242/jcs.230441
143. Senju, Y. *et al.* (2011) Essential role of PACSIN2/syndapin-II in caveolae membrane sculpting. *J. Cell Sci.* 124, 2032–2040
144. Yao, G. *et al.* (2013) The cytoplasmic protein Pacsin 2 in kidney development and injury repair. *Kidney Int.* 83, 426–437
145. Insinna, C. *et al.* (2019) Investigation of F-BAR domain PACSIN proteins uncovers membrane tubulation function in cilia assembly and transport. *Nat. Commun.* 10, 428
146. Rangel, L. *et al.* (2019) Caveolin-1alpha regulates primary cilium length by controlling RhoA GTPase activity. *Sci. Rep.* 9, 1116
147. Jeffries, E.P. *et al.* (2019) Failure to reabsorb the primary cilium induces cellular senescence. *FASEB J.* 33, 4866–4882
148. Hatzfeld, M. *et al.* (2017) Desmosomes and intermediate filaments: their consequences for tissue mechanics. *Cold Spring Harb. Perspect. Biol.* Published online June 1. https://doi.org/10.1101/cshperspect.a029157
149. Giuliodori, A. *et al.* (2018) Loss of cardiac Wnt/beta-catenin signalling in desmoplakin-deficient AC8 zebrafish models is rescuable by genetic and pharmacological intervention. *Cardiovasc. Res.* 114, 1082–1097
150. Chen, S.N. *et al.* (2014) The hippo pathway is activated and is a causal mechanism for adipogenesis in arrhythmogenic cardiomyopathy. *Circ. Res.* 114, 454–468
151. Liu, X. *et al.* (2016) Tead and AP1 coordinate transcription and motility. *Cell Rep.* 14, 1169–1180
152. Davies, P.F. (1995) Flow-mediated endothelial mechanotransduction. *Physiol. Rev.* 75, 519–560
153. Moroishi, T. *et al.* (2015) A YAP/TAZ-induced feedback mechanism regulates Hippo pathway homeostasis. *Genes Dev.* 29, 1271–1284
154. Meng, Z. *et al.* (2016) Mechanisms of Hippo pathway regulation. *Genes Dev.* 30, 1–17
155. Gundogdu, R. and Hergovich, A. (2019) MOB (Mps one Binder) proteins in the Hippo pathway and cancer. *Cells* Published online June 10, 2019. https://doi.org/10.3390/cells8060569
156. Lim, S. *et al.* (2019) Identification of the kinase STK25 as an upstream activator of LATS signaling. *Nat. Commun.* 10, 1547
157. Stegert, M.R. *et al.* (2005) Regulation of NDR protein kinase by hydrophobic motif phosphorylation

mediated by the mammalian Ste20-like kinase MST3. *Mol. Cell. Biol.* 25, 11019–11029
158. Zhang, L. *et al.* (2015) NDR functions as a physiological YAP1 kinase in the intestinal epithelium. *Curr. Biol.* 25, 296–305
159. Chiba, S. *et al.* (2009) MST2- and Furry-mediated activation of NDR1 kinase is critical for precise alignment of mitotic chromosomes. *Curr. Biol.* 19, 675–681
160. Vichalkovski, A. *et al.* (2008) NDR kinase is activated by RASSF1A/MST1 in response to Fas receptor stimulation and promotes apoptosis. *Curr. Biol.* 18, 1889–1895
161. Selimoglu, R.B.A., Joffre, C., Meunier, B. Parrini, M.C. *et al.* (2014) RalA GTPase and MAP4K4 function through NDR1 activation in stress response and apoptotic signaling. *J. Cell Biol. Cell Metab.* Published online August 18, 2014. https://doi.org/10.24966/CBCM-1943/100001
162. Hillmer, R.E. and Link, B.A. (2019) The roles of Hippo signaling transducers Yap and Taz in chromatin remodeling. *Cells* Published online May 24, 2019. https://doi.org/10.3390/cells8050502
163. Li, P. *et al.* (2016) alphaE-catenin inhibits a Src-YAP1 oncogenic module that couples tyrosine kinases and the effector of Hippo signaling pathway. *Genes Dev.* 30, 798–811
164. Si, Y. *et al.* (2017) Src inhibits the Hippo tumor suppressor pathway through tyrosine phosphorylation of Lats1. *Cancer Res.* 77, 4868–4880
165. Robinson, M.S. (2015) Forty years of clathrin-coated vesicles. *Traffic* 16, 1210–1238
166. Yamada, E. (1955) The fine structure of the gall bladder epithelium of the mouse. *J. Biophys. Biochem. Cytol.* 1, 445–458
167. Palade, G.E. (1953) Fine structure of blood capillaries. *J. Appl. Phys.* 24, 1424
168. Stahlhut, M. and van Deurs, B. (2000) Identification of filamin as a novel ligand for caveolin-1: evidence for the organization of caveolin-1-associated membrane domains by the actin cytoskeleton. *Mol. Biol. Cell* 11, 325–337
169. Muriel, O. *et al.* (2011) Phosphorylated filamin A regulates actin-linked caveolae dynamics. *J. Cell Sci.* 124, 2763–2776
170. Sverdlov, M. *et al.* (2009) Filamin A regulates caveolae internalization and trafficking in endothelial cells. *Mol. Biol. Cell* 20, 4531–4540
171. Hansen, C.G. *et al.* (2009) SDPR induces membrane curvature and functions in the formation of caveolae. *Nat. Cell Biol.* 11, 807–814
172. Bastiani, M. *et al.* (2009) MURC/Cavin-4 and cavin family members form tissue-specific caveolar complexes. *J. Cell Biol.* 185, 1259–1273
173. Ludwig, A. *et al.* (2013) Molecular composition and ultrastructure of the caveolar coat complex. *PLoS Biol.* 11, e1001640
174. Sandvig, K. *et al.* (2018) Clathrin-independent endocytosis: an increasing degree of complexity. *Histochem. Cell Biol.* 150, 107–118
175. Guo, Y. *et al.* (2015) Osmotic stress reduces Ca^{2+} signals through deformation of caveolae. *J. Biol. Chem.* 290, 16698–16707
176. Dulhunty, A.F. and Franzini-Armstrong, C. (1975) The relative contributions of the folds and caveolae to the surface membrane of frog skeletal muscle fibres at different sarcomere lengths. *J. Physiol.* 250, 513–539
177. Yeow, I. *et al.* (2017) EHD proteins cooperate to generate caveolar clusters and to maintain caveolae during repeated mechanical stress. *Curr. Biol.* 27, 2951–2962 e2955
178. Hansen, C.G. *et al.* (2011) Pacsin 2 is recruited to caveolae and functions in caveolar biogenesis. *J. Cell Sci.* 124, 2777–2785
179. Hoernke, M. *et al.* (2017) EHD2 restrains dynamics of caveolae by an ATP-dependent, membrane-bound, open conformation. *Proc. Natl. Acad. Sci. U. S. A.* 114, E4360–E4369
180. Moren, B. *et al.* (2012) EHD2 regulates caveolar dynamics via ATP-driven targeting and oligomerization. *Mol. Biol. Cell* 23, 1316–1329
181. Stoeber, M. *et al.* (2012) Oligomers of the ATPase EHD2 confine caveolae to the plasma membrane through association with actin. *EMBO J.* 31, 2350–2364
182. Torrino, S. *et al.* (2018) EHD2 is a mechanotransducer connecting caveolae dynamics with gene transcription. *J. Cell Biol.* 217, 4092–4105
183. Pekar, O. *et al.* (2012) EHD2 shuttles to the nucleus and represses transcription. *Biochem. J.* 444, 383–394
184. Pearse, B.M. (1975) Coated vesicles from pig brain: purification and biochemical characterization. *J. Mol. Biol.* 97, 93–98