Gene Expression **2024** vol. 000(000) | 000–000 Epub
DOI: 10.14218/GE.2023.00192

Review Article

TGF-β and HIPPO Signaling Pathways Interplay in Distinct Hepatic Contexts

Victor M. Color-Aparicio[1], Angeles C. Tecalco-Cruz[2], Blanca Delgado-Coello[1], Marcela Sosa-Garrocho[1], Jaime Mas-Oliva[1], Genaro Vázquez-Victorio[3] and Marina Macías-Silva[1*]

[1]Instituto de Fisiología Celular, Universidad Nacional Autónoma de México, Mexico City, Mexico; [2]Programa en Ciencias Genómicas, Universidad Autónoma de la Ciudad de México, Mexico City, Mexico; [3]Facultad de Ciencias, Universidad Nacional Autónoma de México, Mexico City, Mexico

Received: December 09, 2023 | **Revised:** March 13, 2024 | **Accepted:** May 21, 2024 | **Published online:** June 30, 2024

Abstract

The liver plays a crucial role in maintaining whole-body homeostasis in both health and disease, engaging in important communication with other organs. The coordination of multiple signaling pathways is essential for facilitating such interorgan communication. Among these pathways, the transforming growth factor-β (TGF-β) and HIPPO signaling pathways serve critical functions as tumor suppressors, exerting pivotal control over liver development, size, and tissue regeneration. In the normal hepatic context, these pathways exhibit significant crosstalk through various molecular mechanisms. This interaction is context-dependent within the hepatic microenvironment, regulating diverse cellular processes from development to adulthood. Disruptions in the regulation of these pathways and their crosstalk contribute to the onset of liver diseases. This review delves into the intricate interplay between the canonical pathways of TGF-β and HIPPO, exploring their involvement in liver development and various pathologies such as fibrosis, cirrhosis, and tumorigenesis. Special attention is given to their impact on the epithelial-to-mesenchymal transition process, a crucial element associated with liver wound healing and cancer metastasis. By addressing these molecular interactions, the review aimed to provide insights into the underlying mechanisms that influence liver physiology and pathology, offering potential avenues for therapeutic interventions.

Introduction

Organ intercommunication occurs in multiple physiological and pathological contexts.[1] The liver is one of the main organs that establishes important crosstalk with other organs to maintain whole-body homeostasis, supported by the interplay of multiple signaling pathways.[1] Interestingly, the network of key signaling pathways responsible for orchestrating morphogenesis during embryonic development is reactivated during wound healing and tumorigenesis.[2]

The liver sustains active communication with other organs, which is crucial for the regulation of multiple cellular processes.[1] In liver regeneration, for instance, the liver communicates with other organs, such as the brain, pancreas, intestine, and heart, mainly through cell-cell communication using chemical messengers such as hormones, cytokines, and growth factors.[3] This cellular communication is crucial to restoring hepatic homeostasis after any damage. Among all these signals, the transforming growth factor-β (TGF-β) and HIPPO signaling pathways are particularly relevant, exerting critical functions as tumor suppressors and exhibiting important crosstalk to regulate liver development, size, and regeneration. This review focused on describing the interplay between the TGF-β and HIPPO signaling pathways in distinct hepatic contexts, including the maintenance of liver homeostasis in health, liver regeneration, and the progression of liver diseases such as hepatitis, fibrosis, cirrhosis, and cancer.

Liver physiology and the variety of hepatic contexts in health and disease

The liver stands as a multitasking organ, undertaking crucial functions essential for overall well-being. Among its vital roles, the liver manages the metabolism of nutrients, aids in digestion by producing bile for fat digestion, and actively participates in the detoxification of blood by processing toxins, drugs, and viruses. Additionally, the liver synthesizes proteins for blood plasma, regulates blood clotting, and serves as a frontline immune tissue, contributing significantly to the body's immune response.

Keywords: TGF-β; SMAD; HIPPO; TAZ; YAP; Liver.

***Correspondence to:** Marina Macías-Silva, Departamento de Biología Celular y Desarrollo, Instituto de Fisiología Celular, Universidad Nacional Autónoma de México, Ciudad Universitaria, Coyoacán, Ciudad de México, Mexico City, C.P. 04510, Mexico. ORCID: https://orcid.org/0000-0002-3972-0983. Tel: +52-55-56225729, E-mail: mmacias@ifc.unam.mx

How to cite this article: Color-Aparicio VM, Tecalco-Cruz AC, Delgado-Coello B, Sosa-Garrocho M, Mas-Oliva J, Vázquez-Victorio G, *et al*. TGF-β and HIPPO Signaling Pathways Interplay in Distinct Hepatic Contexts. *Gene Expr* 2024;000(000):000–000. doi: 10.14218/GE.2023.00192.

The hepatic lobules are the functional units of the liver, populated by different cell types that maintain constant intercellular communication through various signaling pathways in different contexts. Hepatocytes are the most abundant parenchymal cell type (80%) in the liver, whereas the non-parenchymal cells (20%) include hepatic stellate cells (HSCs), Kupffer cells, liver sinusoidal endothelial cells (LSECs), lymphocytes, and biliary epithelial cells (BECs) or cholangiocytes.[3] The hepatic tissue is crucial during all stages of organism development and shows relevant differences regarding hepatic cell proliferation and function in early embryonic stages compared to adulthood. For instance, in adulthood, the liver is an organ whose cells rarely divide, remaining quiescent most of the time; it is estimated that one cell among 20,000 cells undergoes mitosis.[3]

Contrary to what is observed in the adult mammalian liver, where hepatic cells are typically quiescent, fetal hepatoblasts migrate together as cords while inserting between mesenchymal and epithelial cells to form the primitive liver. Hepatoblasts are bipotential cells that actively proliferate and differentiate into hepatocytes and cholangiocytes.[4] Once the hepatic vasculature is constructed, the fetal liver performs hematopoiesis around 10.5–12.5 days post-fertilization. Despite the technical issues involved in studying embryonic stages, we have a good understanding of the main phases involved in early liver development (progenitor specification, cell type-specific differentiation, and outgrowth) and the role of the placenta in providing oxygen, calcium, and possibly bile acids.[2,4]

An amazing property of the liver in adult vertebrates is its ability to regenerate. After exposure to stimuli provoking the loss of hepatic mass (e.g., the 70% partial hepatectomy (PH) model) or tissue damage, a phenomenon of rapid recovery of the original liver size occurs through a compensatory hyperplasia mechanism along with restoration of hepatic architecture and functions, known as liver regeneration (Fig. 1). Interestingly, other organs can undergo regeneration but not at the same scale as the liver, whose size is strictly regulated through an intricate signaling network. This molecular toolkit, evolutionarily acquired and improved in mammals,[3] includes pro-inflammatory and inhibitory cytokines, growth factors, members of different signaling pathways and transcription factors, as well as diverse epigenetic mechanisms. Together, they participate in the activation and deactivation of genes in a highly regulated manner.

It is important to note that all hepatic cell types contribute to recovering the liver mass and express outstanding functional and phenotypic changes. For instance, during the activation process of HSCs, these cells lose vitamin A storage, acquire a contractile myofibroblast-like phenotype, and synthesize distinct collagen types. Kupffer cells produce pro-inflammatory cytokines to promote changes in the hepatocytes that turn them responsive to growth factors. During liver regeneration after a 70% PH, all hepatic cells are activated by cytokines (e.g., interleukin-6 (IL-6) and tumor necrosis factor-alpha) and growth factors (e.g. hepatocyte growth factor and epidermal growth factor) that promote the overexpression of specific genes, allowing the cells to enter the cell cycle and go through each stage of regeneration (priming, proliferative, and termination phases). This process takes approximately seven days in rats until the mass, architecture and function of the liver are restored. All liver cells contribute to the process, with hepatocytes being the first to divide, followed by BECs, Kupffer cells, HSCs, and finally LSECs to complete the reconstruction of extracellular matrix (ECM) architecture and hepatic vasculature.[3]

Significantly, when the regenerative capabilities of the liver are surpassed, alternative scenarios unfold wherein liver cells manifest altered phenotypes in response to chemical or physical damage (Fig. 1). For instance, HSCs undergo prolonged activation with sustained exposure to damage from drugs, alcohol, or a high-fat diet. As a result, HSCs adopt a myofibroblast-like phenotype, leading to excessive production of ECM proteins, forming a perfect microenvironment for the development of fibrosis and cirrhosis, eventually leading to liver failure and, in some cases, hepatocarcinogenesis. The progression of fibrosis involves the gradual replacement of parenchymal tissue with scar tissue, altering the hepatic architecture and increasing stiffness. In the resolution stage of fibrosis, the liver returns to homeostasis, and the activated HSCs can be eliminated via apoptosis promoted by macrophages with pro-resolution properties.[5] However, if the liver damage is chronic, cirrhosis becomes a point of no return, characterized by severe scarring of the liver and significantly increasing the risk of developing hepatocellular carcinoma (HCC) (Fig. 1). Liver cancer, one of the deadliest cancers worldwide, primarily manifests as two types: HCC (malignant transformation of hepatocytes) and intrahepatic cholangiocarcinoma (ICC) (cancer of BECs).

In the aforementioned hepatic contexts, numerous signaling pathways assume pivotal roles. Therefore, understanding the intricate networks of signaling pathways contributing to the progression of liver disease is crucial for identifying potential therapeutic targets.[6] Given the significant impact chronic liver diseases have worldwide, we have recently reviewed various interdisciplinary efforts aimed at mitigating this trend.[7] In extreme cases of liver damage where patients may need a liver transplant, the situation is exacerbated by the low number of donors. Thus, options for patients with acute liver failure or severe chronic liver diseases include extracorporeal liver support devices that function while waiting for an organ donor. Another alternative provided by tissue engineering is the "recycling" of ECM obtained after liver decellularization for repopulation with healthy liver cells (bioengineered liver), which can eventually be transplanted.[7] Recently, Hans Clever's group has provided a promising alternative for personalized medicine, demonstrating that human and mouse hepatocytes can be cultured long-term as organoids with the potential to engraft and proliferate in a damaged liver.[8] Targeting key signaling pathways has been another important strategy to improve liver regeneration and restore homeostasis in hepatic diseases. Nevertheless, the main drawback of inhibiting key pathways is the potential cytotoxic side effects on normal cells.

Actions of TGF-β and HIPPO signaling pathways in the liver

The HIPPO and TGF-β pathways exert crucial functions in hepatic physiology by regulating essential cellular processes such as proliferation, differentiation, and apoptosis. These pathways control liver mass, architecture, and function from embryonic development through adulthood. The HIPPO and TGF-β signaling pathways have pleiotropic effects, and any gain or loss of their function, as well as aberrant actions, can lead to diseases such as chronic liver inflammation, fibrosis, and cancer.[9–11]

TGF-β is the prototype of a family of multifunctional polypeptides, including TGF-βs, ACTIVINs, INHIBINs, and BMPs (bone morphogenetic proteins). TGF-β initiates signaling through a complex of two types of transmembrane Ser/Thr kinase receptors, TβRII and TβRI (or activin-like kinase 5), which transduce signals via downstream effectors named (receptor-regulated SMADs) R-SMADs, such as SMAD2 and SMAD3. After phosphorylation by the type I receptor, these proteins form complexes with the common-SMAD4 for their translocation to the nucleus, where the SMAD2/3/4 complex binds to DNA sequences (tandem repeats of GTCT or AGAC), known as SMAD-binding elements (SBE) located on TGF-β target genes to tightly control their expression (Fig. 2).[12,13]

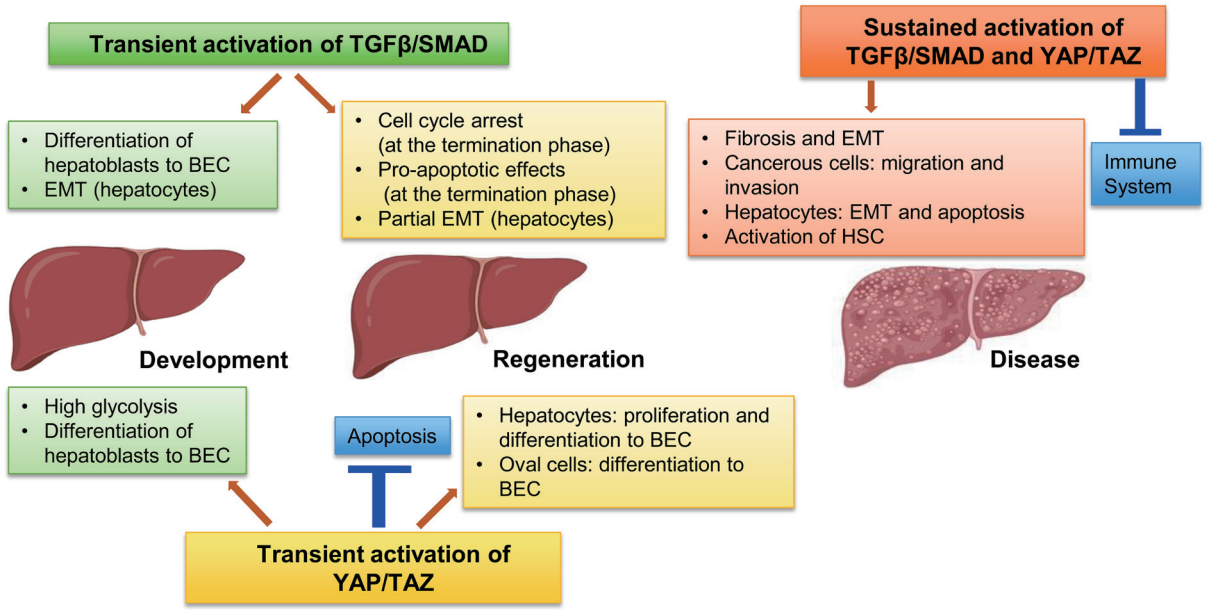

Fig. 1. The roles of HIPPO and TGF-β signaling pathways play in distinct hepatic contexts present in health and disease. BEC, biliary epithelial cells; EMT, epithelial-mesenchymal transition; HSC, hepatic stellate cells; TAZ, transcriptional co-activator with PDZ-binding motif; TGF-β, transforming growth factor-beta; YAP, Yes-associated protein.

TGF-β signaling is negatively regulated by multiple factors. For instance, the Sloan Kettering Institute (SKI) and SKI-novel (SNON) proteins are transcriptional cofactors that function as negative regulators of TGF-β signaling by associating with SMAD proteins to actively turn off the TGF-β canonical pathway through negative feedback mechanisms.[6,14] The TGF-β/SMAD signaling pathway becomes activated during murine liver regeneration, with SKI and SNON proteins being upregulated to modulate the magnitude and

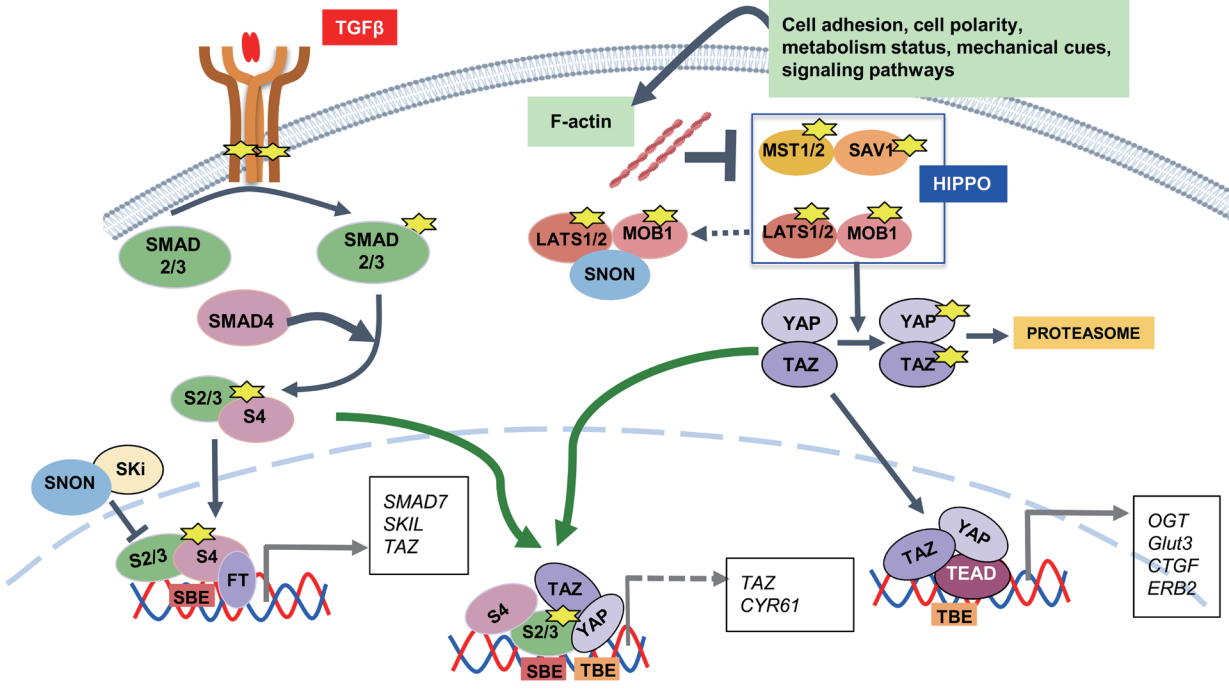

Fig. 2. The canonical HIPPO and TGF-β signaling pathways. CYR61, cysteine-rich angiogenic inducer 61; FT, factors of transcription; LATS, large tumor suppressor; MOB, mob kinase activator; MST, mammalian sterile 20-like kinase; SBE, SMAD-binding elements; SKI, Sloan Kettering Insitute; SKIL, SKI-like; SNON, SKI-novel; TAZ , transcriptional co-activator with PDZ-binding motif; TBE, TEAD-binding elements; TEAD, transcriptional enhanced associate domain; TGF-β, transforming growth factor-beta; YAP, Yes-associated protein.

duration of the TGF-β/SMAD pathway functions in this context (Fig. 2).[15–17] Additionally, evidence shows that SNON/SKI protein stability is differentially regulated by actin cytoskeleton dynamics in hepatic cells, similar to how YAP/TAZ (Yes-associated protein 1/transcriptional co-activator with PDZ-binding motif) stability is controlled by F-ACTIN cytoskeleton rearrangements. Intriguingly, there is evidence that SNON promotes TAZ protein stabilization and signaling in human breast cancer by sequestering LATS kinase.[18] Given the relevance of HIPPO/TGF-β crosstalk in liver regeneration, this positive regulation of TAZ by SNON deserves investigation in the hepatic context.

In liver cancer, the tumor microenvironment dictates that TGF-β functions as a tumor suppressor in early stages but becomes a tumor promoter in advanced stages.[19] Snorri Thorgeirson's group reported two main TGF-β-dependent gene expression signatures in mouse hepatocytes that are useful for predicting clinical outcomes in human liver cancer and distinguishing between subgroups of HCC.[20] An early TGF-β signature (high expression of anti-proliferative and anti-apoptotic genes) correlated with a good prognosis for patients with liver cancer, whereas a late TGF-β signature (high expression of invasion-related, epithelial-mesenchymal-transition (EMT), and anti-apoptotic genes) was associated with tumor recurrence and metastasis.[20,21]

The HIPPO signaling pathway is regulated by diverse upstream signals such as cell polarity, cell adhesions, mechanical forces, cytoskeleton dynamics, G protein coupled receptors (GPCR) ligands, and some stress signals.[22] The HIPPO pathway comprises two core kinases, mammalian sterile 20-like kinase 1 (MST1) and MST2, which activate large tumor suppressor 1 (LATS1) and LATS2 kinases through phosphorylation.[10,23,24] A cytoskeletal protein named NF2/moesin-ezrin-radixin like (MERLIN) regulates MST1/2 kinases and their adaptor protein SAV1/WW45, while activators MOB kinase activator 1A (MOB1A) and MOB1B regulate LATS1/2. The main effectors of the HIPPO signaling pathway are the transcriptional co-regulators TAZ/WW domain-containing transcription regulator 1 (WWTR1) and YAP, which are negatively regulated by HIPPO signaling (Fig. 2). In the liver, signals like mechanical forces generated by the F-ACTIN cytoskeleton and high energy levels may negatively regulate HIPPO signaling. When the HIPPO pathway is active, the LATS1 and LATS2 kinases phosphorylate YAP and TAZ, leading to their inactivation. Other post-transcriptional modifications can modulate YAP/TAZ protein stability and activity, such as ubiquitination, methylation, acetylation, and O-GlcNAcylation.[25] YAP/TAZ inactivation might occur through several mechanisms, such as their phosphorylation resulting in exclusion from the nucleus, or their ubiquitination promoting degradation via the proteasome.[10,26] When the HIPPO pathway is inactive, MST1/2 and LATS1/2 kinases are inactivated, allowing YAP and TAZ to become active and translocate to the nucleus to regulate gene expression. YAP and TAZ are ubiquitously expressed transcriptional cofactors that share similar structures and functions and may be regulated by similar mechanisms, although they have different roles from development to adulthood. Notably, YAP/TAZ co-regulators can interact with many other transcription factors besides transcriptional enhanced associate domain (TEADs), such as RUNX2 (runt-related transcription factor 2), TBX5 (T-box transcription factor 5), P73, and SMADs, allowing for the control of specific context-dependent gene expression (Fig. 2).[10] Thus, they can regulate the expression of target genes related to several cellular processes, such as proliferation, differentiation, and EMT.[10,27–29]

In the liver, the HIPPO pathway is one of the main signaling pathways sustaining hepatic physiology. It plays a crucial role in maintaining the quiescence of hepatic cells and contributes to proper metabolic zonation of the liver.[30,31] Consequently, deregulation of the HIPPO pathway, along with other factors, has been associated with metabolic diseases such as type 2 diabetes, and non-alcoholic fatty liver disease. Moreover, overexpression of YAP and TAZ in the liver promotes cell proliferation that may lead to hepatomegaly and eventually hepatocarcinogenesis, revealing an outstanding role in regulating organ size in various scenarios.[27,32] For instance, during liver regeneration, HIPPO signaling becomes relevant for controlling mouse liver size, as shown in the conditional liver double knockout of *YAP* and *TAZ*, where regeneration is impaired, causing a longer restoration period for liver mass.[27,33] In contrast, studies in young mice show a redundancy of other pathways that permit liver regeneration to take place but at a slower rate.[34] When mice livers are exposed to chronic damage, liver repair is associated with fibrosis, and in this context, HSCs are activated for extended periods, producing excess ECM proteins. YAP is activated during HSC activation triggered by different stimuli and is associated with increased ECM protein deposition in the long term. Meanwhile, other non-parenchymal cells, such as LSEC, also show YAP activation, promoting angiogenesis and thereby increasing mice liver damage.[35] In these hepatic contexts, TGF-β and HIPPO pathways converge to regulate organ size, regeneration, and fibrosis, but the mechanisms involved require further studies.

Furthermore, genetic alterations of some HIPPO signaling components, such as knockouts (*NF2*, *SAV1*/*WW45*, or *MST1*/*2*) or overexpressions (*YAP* or *TAZ*), promote liver overgrowth and tumorigenesis.[30,36–41] Although DNA mutations in the main HIPPO pathway components are rare, irregular activation of YAP/TAZ in liver cancer has been described, although the molecular mechanisms involved remain uncertain.[10,41–46] In liver cancer, YAP/TAZ activation can also be regulated by many extrinsic signals, such as growth factors, cytokines, stress signals, altered metabolic conditions, autophagy, and mechanical forces.[47–55] Intriguingly, YAP/TAZ also cooperate with different signaling pathways in a context-dependent manner, such as TGF-β, RTK/PI3K, WNT, and NOTCH signaling.[56–60]

Crosstalk between TGF-β and HIPPO signaling pathways in distinct hepatic contexts

A major crosstalk between the HIPPO and TGF-β pathways occurs to control liver size and regeneration, while their deregulation promotes fibrosis and hepatocarcinogenesis. It is well known that both pathways inhibit cell proliferation and maintain hepatocyte homeostasis, acting primarily as tumor suppressors.[19,57,61] HIPPO downstream effectors, such as TEAD and TAZ/YAP, can function as cofactors of the TGF-β/SMAD canonical pathway, since some gene promoters contain both SBE and TBE (TEAD-binding elements) (Fig. 3). When HIPPO signaling is inactive, dephosphorylated YAP and TAZ proteins translocate to the nucleus, where they associate with members of the TEAD transcription factor family to bind target gene promoters, inducing the expression of genes involved in cell proliferation and inhibition of apoptosis.[62] In the context of low cell density, the HIPPO pathway is likewise inactive, allowing YAP/TAZ to facilitate the accumulation of SMAD proteins in the nucleus in response to TGF-β. Consequently, SMAD/YAP/TAZ complexes can synergize transcriptionally (Fig. 3).[57,60,61,63–65] In contexts such as breast cancer cells, where TGF-β/SMAD signaling is active, this pathway may cooperate with YAP/

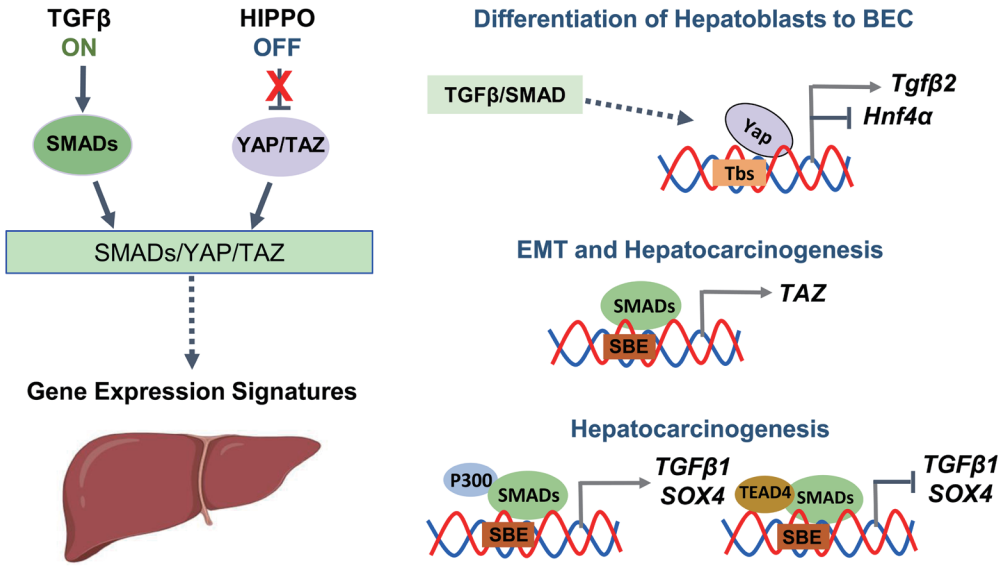

Fig. 3. Gene expression is regulated by the HIPPO/YAP/TAZ and TGF-β/SMAD crosstalk in the liver. BEC, biliary epithelial cells; EMT, Epithelial-Mesenchymal Transition; HNF4α, hepatocyte nuclear factor alpha; SBE, SMAD-binding elements; TAZ, transcriptional co-activator with PDZ-binding motif; Tbs, TEAD binding sites; TGF-β, transforming growth factor-beta; YAP, Yes-associated protein.

TAZ to induce fibrogenic factors, such as *CTGF*, *NEGR1*, and *UCA1*, as well as genes encoding ECM proteins.[66] Therefore, the close relationship between TGF-β and HIPPO pathways in the development of fibrosis and cirrhosis merits further studies.

The involvement of the HIPPO and TGF-β pathways in tumorigenesis has been extensively investigated. It is well-established that the interplay between these pathways can induce the EMT process.[67] EMT is a key hallmark of cancer related to metastasis, although it also becomes activated during development and wound healing. In this process, epithelial cells can completely or partially transdifferentiate (partial EMT), losing epithelial markers and acquiring characteristics similar to mesenchymal cells. This is crucial for cancerous cells, as EMT confers features such as resistance to apoptosis, drug resistance, cancer stem cell characteristics, and invasive abilities.[68,69] Recently, it was shown that the *TGFB1* gene is overexpressed in the murine AS-30D hepatocarcinoma, and more importantly, treatment of these cells with TGF-β increased the levels of phospho-SMAD2 and the expression of several mesenchymal markers.[70] Given the potential of AS-30D cells to undergo EMT, the plausible interplay between the TGF-β and HIPPO signaling pathways in this HCC model is evident.

EMT is regulated by different signals depending on the context. For instance, TGF-β signaling may cooperate with YAP/TAZ to induce EMT in liver cancer.[67,70] The expression of *TAZ* is enhanced in cancer cells from brain, breast, pancreas, and liver tumors,[42,71–74] and appears to mediate cancer stem cell characteristics promoted by TGF-β/SMADs.[60,63,64] In liver cancer, YAP and TAZ promote cellular proliferation, stem cell marker expression, and EMT.[73,75–77] In fact, EMT may serve as a self-sustaining mechanism for TAZ expression and activation.[74] The molecular mechanisms regulating *TAZ/WWTR1* gene expression are poorly studied. Thus far, the *TAZ/WWTR1* gene is regulated by several transcriptional factors such as STAT3, SMAD3, MRTF, and HIF1, whereas different miRNAs regulate *TAZ* mRNA translation. Recent evidence shows that TGF-β cytokine uses distinct molecular mechanisms to control *TAZ/WWTR1* gene expression in a context-dependent manner: Firstly, a non-canonical TGF-β pathway can increase TAZ levels in murine fibroblasts and some pig epithelial cells in a P38/MRTF-dependent and SMAD3-independent manner.[78] Secondly, the synergistic action of IL-6 and TGF-β pathways is essential for inducing *TAZ/WWTR1* gene expression in mouse CD4+ lymphocytes, leading to their differentiation into Th17 cells. This phenomenon is intriguing given that TAZ deficiency promotes the differentiation of lymphocytes toward regulatory T cells (Treg).[79] Notably, the TGF-β pathway regulates *TAZ/WWTR1* gene expression depending on the cellular context and species-specific gene promoter. This evidence suggests that TGF-β predominantly controls *TAZ/WWTR1* gene expression through Smad-independent pathways or in collaboration with other signaling pathways such as IL-6/STAT3.[78,79]

There is evidence in human HCC samples of a correlation between the upregulation of TGF-β and HIPPO pathway components with increased levels of EMT markers, as shown by analyzing LIHC public datasets.[80,81] In contrast to previous reports, we recently described that the TGF-β/SMAD pathway promotes human *TAZ/WWTR1* gene expression and enhances TAZ protein levels in human cancer hepatic cells. Thus, TAZ is a primary target of TGF-β signaling, a major pathway in hepatic cancer development.[80] Moreover, our investigation revealed that the human *TAZ/WWTR1* gene promoter encompasses both canonical and non-canonical SBEs. Particularly noteworthy is the identification of a TRE (TGF-β-response element) within the *TAZ/WWTR1* gene promoter, formed by canonical SBEs.[80] Consequently, our findings indicate that the TGF-β/SMAD canonical pathway plays a regulatory role in TAZ expression within human hepatic cancer cells by amplifying *TAZ/WWTR1* gene transcription and enhancing TAZ protein stability (Fig. 3). Our findings suggest that TGF-β/SMAD signaling might cooperate with TAZ in liver cancer progression, probably through the regulation of a specific gene signature. Furthermore, the conflicting evidence suggesting that TGF-β regulates *TAZ/WWTR1* gene expression through SMAD or non-SMAD signaling needs further investigation to elucidate the relevance of the differential regulation of *TAZ/WWTR1* gene expression depending on the cellular context and species-specific gene promoter.

The crosstalk between TGF-β and HIPPO pathways is crucial in various hepatic contexts, from embryonic development to homeostasis and disease in adulthood. The interplay between these pathways may generate a TGF-β/SMAD/YAP/TAZ axis that likely controls specific gene expression networks depending on the different hepatic contexts present in liver health or disease (Fig. 3). Studies on mouse embryo development show that TGF-β and HIPPO pathways cooperate to promote mesoderm to endoderm differentiation during liver formation and maturation; in this context, YAP activation in hepatoblasts promotes increased TGF-β signaling, leading to hepatoblast differentiation into BECs.[2,11,31] Moreover, some murine models, such as mice liver-conditional KO of *LATS1/2* kinases are used to study the fate of hepatic progenitors.[82] In BECs obtained from these livers, the loss of *LATS1/2* promotes SMAD2/3 nuclear localization and the binding of YAP to the mouse *TGFB2* gene promoter, resulting in the upregulation of genes such as *TGFB2*, *CK7/KRT7*, and *CK9/KRT9*. YAP also promotes the downregulation of HNF4α (Hepatocyte Nuclear Factor 4 alpha) by binding to the mouse *HNF4a* gene promoter, influencing cells fate determination by inducing hepatoblast differentiation into BECs.[82] This model demonstrates the crosstalk between HIPPO and TGF-β signaling pathways in the differentiation of mouse hepatoblasts into BECs.[82–84]

Liver regeneration is a primary context where the interplay between TGF-β and HIPPO pathways is evident. After PH in mice, the nuclear localization of SMAD2 and YAP increases in proliferating hepatocytes undergoing partial EMT, correlating with the upregulation of *SNAI1*, *ZEB1*, *aSMA*, *COL1a1*, *VIM*, *MMP9,* and *QSOX1* genes, and the downregulation of *CDH1* (E-cadherin) and *ALB* genes. In this model, TGF-β and YAP1 cooperate to induce partial EMT in hepatocytes, which acquire fibroblast-like characteristics, allowing hepatocytes to proliferate and manage anti-proliferative signals present in the microenvironment.[67] In a human cell line of HSCs, *TAZ* overexpression induces the upregulation of genes associated with fibrosis, such as *TGFB1* and *SNAI1*.[85] In a rat model of hepatic fibrosis induced by diethyl-nitrosamine, the nuclear co-localization of SMAD2/3 and YAP/TAZ increases, along with the upregulation of MMP2, MMP9, and TIMP1 proteins, and the downregulation of MMP1 protein.[86] Additionally, in human hepatic fibrosis, TGF-β can promote the activation of PYK2 (proline-rich tyrosine kinase 2), while SRC (Sarcoma kinase) activates the RHOA/ROCK axis, increasing YAP nuclear localization in an HSC cell line. Together, TGF-β and HIPPO pathways cooperate to upregulate genes such as *CTGF* and *CYR61* in activated human HSC cell line.[87] Intriguingly, SIRT6, a NAD-dependent deacetylase, has therapeutic potential as it protects against liver fibrosis by deacetylating key lysine residues on SMAD2, SMAD3, YAP, and TAZ, causing their inactivation.[88–90] Thus, liver regeneration benefits from the interplay between TGF-β and HIPPO pathways.

The main types of liver cancer, ICC and HCC, are induced in the *MOB1a*/*MOB1b*-deficient mouse model. In this liver cancer model, there is an observed increase in the nuclear co-localization of SMAD2 and YAP/TAZ, along with increased levels of CTGF, TGF-β2, and TGF-β3 protein levels.[91] Moreover, the upregulation of YAP, SMAD2/3, and TGF-β2, combined with the downregulation of MOB1, may serve as a prognostic indicator of poor survival for ICC patients.[92] In human HCC cells, TGF-β promotes an increase in the nuclear localization of the SMAD2/3 complex, which can recruit P300 to promote the expression of *TGFB1* and *SOX4* genes, associated with increased proliferation, migration, and invasion of HCC cells. Intriguingly, the ectopic expression of TEAD4 in these HCC cells allows TEAD4 to associate with SMAD2/3/4, competing with P300 to repress *TGFB1* and *SOX4* gene expression, through a YAP/TAZ-independent mechanism. This crosstalk between TGF-β/SMADs and HIPPO/TEAD4 in cancer is relevant as it inhibits specific gene expression signatures related to HCC progression.[93]

Conclusions

The interplay between the HIPPO and TGF-β canonical pathways plays a significant role in various hepatic contexts. In certain scenarios, the inactivation of the HIPPO pathway leads to the activation of its principal downstream effectors, the transcriptional cofactors YAP and TAZ. The activation of these cofactors can intersect with the activation of the TGF-β/SMAD pathway, forming an axis termed SMAD/YAP/TAZ. This axis effectively governs specific gene expression signatures that may contribute to diverse physiological or pathological processes in the liver. Therefore, it is imperative to identify these genetic signatures to delineate potential targets for therapeutic interventions.

Acknowledgments

We thank all members of Unidad de Imagenología and Bioterio at Instituto de Fisiología Celular, UNAM.

Funding

This work was supported in part by a grant from the Consejo Nacional de Humanidades, Ciencias y Tecnologías (No. 304023 to GVV and MMS). VMCA received a postdoctoral fellowship from the Consejo Nacional de Humanidades, Ciencias y Tecnologías (grant No. 304023).

Conflict of interest

The authors have no conflict of interest related to this publication.

Author contributions

Conceptualization of the review (MMS); collection of information (VMCA, ACTC, BDC, MSG, JMO, GVV, MMS); writing of original draft (VMCA, ACTC, BDC, MMS); figures (VMCA, MSG); intellectual input, critical manuscript revisions, and final draft approval (all authors).

References

[1] Wang F, So KF, Xiao J, Wang H. Organ-organ communication: The liver's perspective. Theranostics 2021;11(7):3317–3330. doi:10.7150/thno.55795, PMID:33537089.

[2] Campbell SA, Stephan TL, Lotto J, Cullum R, Drissler S, Hoodless PA. Signalling pathways and transcriptional regulators orchestrating liver development and cancer. Development 2021;148(17):dev199814. doi:10.1242/dev.199814, PMID:34478514.

[3] Michalopoulos GK, Bhushan B. Liver regeneration: biological and pathological mechanisms and implications. Nat Rev Gastroenterol Hepatol 2021;18(1):40–55. doi:10.1038/s41575-020-0342-4, PMID:32764740.

[4] Lotto J, Stephan TL, Hoodless PA. Fetal liver development and implications for liver disease pathogenesis. Nat Rev Gastroenterol Hepatol 2023;20(9):561–581. doi:10.1038/s41575-023-00775-2, PMID:37208503.

[5] Iredale JP, Thompson A, Henderson NC. Extracellular matrix degrada-

tion in liver fibrosis: Biochemistry and regulation. Biochim Biophys Acta 2013;1832(7):876–883. doi:10.1016/j.bbadis.2012.11.002, PMID:23149387.

[6] Tecalco-Cruz AC, Sosa-Garrocho M, Vázquez-Victorio G, Ortiz-García L, Domínguez-Hüttinger E, Macías-Silva M. Transforming growth factor-β/SMAD Target gene SKIL is negatively regulated by the transcriptional cofactor complex SNON-SMAD4. J Biol Chem 2012;287(32):26764–26776. doi:10.1074/jbc.M112.386599, PMID:22674574.

[7] Delgado-Coello B, Navarro-Alvarez N, Mas-Oliva J. The Influence of Interdisciplinary Work towards Advancing Knowledge on Human Liver Physiology. Cells 2022;11(22):3696. doi:10.3390/cells11223696, PMID:36429123.

[8] Hu H, Gehart H, Artegiani B, LÖpez-Iglesias C, Dekkers F, Basak O, *et al*. Long-Term Expansion of Functional Mouse and Human Hepatocytes as 3D Organoids. Cell 2018;175(6):1591–1606.e19. doi:10.1016/j.cell.2018.11.013, PMID:30500538.

[9] Yimlamai D, Fowl BH, Camargo FD. Emerging evidence on the role of the Hippo/YAP pathway in liver physiology and cancer. J Hepatol 2015;63(6):1491–1501. doi:10.1016/j.jhep.2015.07.008, PMID:26226451.

[10] Zanconato F, Cordenonsi M, Piccolo S. YAP/TAZ at the Roots of Cancer. Cancer Cell 2016;29(6):783–803. doi:10.1016/j.ccell.2016.05.005, PMID:27300434.

[11] Fabregat I, Caballero-Díaz D. Transforming Growth Factor-β-Induced Cell Plasticity in Liver Fibrosis and Hepatocarcinogenesis. Front Oncol 2018;8:357. doi:10.3389/fonc.2018.00357, PMID:30250825.

[12] Vázquez-Victorio G, Rosales-Alvarez RE, Ríos-López DG, Tecalco-Cruz AC, Macías-Silva M. Chapter 4: TGF-beta signaling pathway regulation by transcriptional cofactors Ski and SnoN in health and disease. In: Duncan LT (ed). Advances in Health and Disease. 1st ed, Vol 1. New York: NOVA Science Publishers Inc; 2017:1–74.

[13] David CJ, Massagué J. Publisher Correction: Contextual determinants of TGFβ action in development, immunity and cancer. Nat Rev Mol Cell Biol 2018;19(7):479. doi:10.1038/s41580-018-0018-x, PMID:29740128.

[14] Tecalco-Cruz AC, Ríos-López DG, Vázquez-Victorio G, Rosales-Alvarez RE, Macías-Silva M. Transcriptional cofactors Ski and SnoN are major regulators of the TGF-β/Smad signaling pathway in health and disease. Signal Transduct Target Ther 2018;3:15. doi:10.1038/s41392-018-0015-8, PMID:29892481.

[15] Macias-Silva M, Li W, Leu JI, Crissey MA, Taub R. Up-regulated transcriptional repressors SnoN and Ski bind Smad proteins to antagonize transforming growth factor-beta signals during liver regeneration. J Biol Chem 2002;277(32):28483–28490. doi:10.1074/jbc.M202403200, PMID:12023281.

[16] Vázquez-Victorio G, Caligaris C, Del Valle-Espinosa E, Sosa-Garrocho M, González-Arenas NR, Reyes-Cruz G, *et al*. Novel regulation of Ski protein stability and endosomal sorting by actin cytoskeleton dynamics in hepatocytes. J Biol Chem 2015;290(7):4487–4499. doi:10.1074/jbc.M114.579532, PMID:25561741.

[17] Caligaris C, Vázquez-Victorio G, Sosa-Garrocho M, Ríos-López DG, Marín-Hernández A, Macías-Silva M. Actin-cytoskeleton polymerization differentially controls the stability of Ski and SnoN co-repressors in normal but not in transformed hepatocytes. Biochim Biophys Acta 2015;1850(9):1832–1841. doi:10.1016/j.bbagen.2015.05.012, PMID:26002202.

[18] Zhu Q, Le Scolan E, Jahchan N, Ji X, Xu A, Luo K. SnoN Antagonizes the Hippo Kinase Complex to Promote TAZ Signaling during Breast Carcinogenesis. Dev Cell 2016;37(5):399–412. doi:10.1016/j.devcel.2016.05.002, PMID:27237790.

[19] Gungor MZ, Uysal M, Senturk S. The Bright and the Dark Side of TGF-β Signaling in Hepatocellular Carcinoma: Mechanisms, Dysregulation, and Therapeutic Implications. Cancers (Basel) 2022;14(4):940. doi:10.3390/cancers14040940, PMID:35205692.

[20] Coulouarn C, Factor VM, Thorgeirsson SS. Transforming growth factor-beta gene expression signature in mouse hepatocytes predicts clinical outcome in human cancer. Hepatology 2008;47(6):2059–2067. doi:10.1002/hep.22283, PMID:18506891.

[21] Fabregat I, Giannelli G, IT-LIVER Consortium. The TGF-β pathway: a pharmacological target in hepatocellular carcinoma? Hepat Oncol 2017;4(2):35–38. doi:10.2217/hep-2017-0012, PMID:30191051.

[22] Zheng Y, Pan D. The Hippo Signaling Pathway in Development and Disease. Dev Cell 2019;50(3):264–282. doi:10.1016/j.devcel.2019.06.003, PMID:31386861.

[23] Yagi R, Chen LF, Shigesada K, Murakami Y, Ito Y. A WW domain-containing yes-associated protein (YAP) is a novel transcriptional co-activator. EMBO J 1999;18(9):2551–2562. doi:10.1093/emboj/18.9.2551, PMID:10228168.

[24] Kanai F, Marignani PA, Sarbassova D, Yagi R, Hall RA, Donowitz M, *et al*. TAZ: a novel transcriptional co-activator regulated by interactions with 14-3-3 and PDZ domain proteins. EMBO J 2000;19(24):6778–6791. doi:10.1093/emboj/19.24.6778, PMID:11118213.

[25] Zhang S, Zhou D. Role of the transcriptional coactivators YAP/TAZ in liver cancer. Curr Opin Cell Biol 2019;61:64–71. doi:10.1016/j.ceb.2019.07.006, PMID:31387016.

[26] Hao Y, Chun A, Cheung K, Rashidi B, Yang X. Tumor suppressor LATS1 is a negative regulator of oncogene YAP. J Biol Chem 2008;283(9):5496–5509. doi:10.1074/jbc.M709037200, PMID:18158288.

[27] Su T, Bondar T, Zhou X, Zhang C, He H, Medzhitov R. Two-signal requirement for growth-promoting function of Yap in hepatocytes. Elife 2015;4:e02948. doi:10.7554/eLife.02948, PMID:25667983.

[28] Weiler SME, Lutz T, Bissinger M, Sticht C, Knaub M, Gretz N, *et al*. TAZ target gene ITGAV regulates invasion and feeds back positively on YAP and TAZ in liver cancer cells. Cancer Lett 2020;473:164–175. doi:10.1016/j.canlet.2019.12.044, PMID:31904487.

[29] Marquard S, Thomann S, Weiler SME, Bissinger M, Lutz T, Sticht C, *et al*. Yes-associated protein (YAP) induces a secretome phenotype and transcriptionally regulates plasminogen activator Inhibitor-1 (PAI-1) expression in hepatocarcinogenesis. Cell Commun Signal 2020;18(1):166. doi:10.1186/s12964-020-00634-6, PMID:33097058.

[30] Molina LM, Zhu J, Li Q, Pradhan-Sundd T, Krutsenko Y, Sayed K, *et al*. Compensatory hepatic adaptation accompanies permanent absence of intrahepatic biliary network due to YAP1 loss in liver progenitors. Cell Rep 2021;36(1):109310. doi:10.1016/j.celrep.2021.109310, PMID:34233187.

[31] Nguyen-Lefebvre AT, Selzner N, Wrana JL, Bhat M. The hippo pathway: A master regulator of liver metabolism, regeneration, and disease. FASEB J 2021;35(5):e21570. doi:10.1096/fj.202002284RR, PMID:33831275.

[32] Patel SH, Camargo FD, Yimlamai D. Hippo Signaling in the Liver Regulates Organ Size, Cell Fate, and Carcinogenesis. Gastroenterology 2017;152(3):533–545. doi:10.1053/j.gastro.2016.10.047, PMID:28003097.

[33] Lu L, Finegold MJ, Johnson RL. Hippo pathway coactivators Yap and Taz are required to coordinate mammalian liver regeneration. Exp Mol Med 2018;50(1):e423. doi:10.1038/emm.2017.205, PMID:29303509.

[34] Loforese G, Malinka T, Keogh A, Baier F, Simillion C, Montani M, *et al*. Impaired liver regeneration in aged mice can be rescued by silencing Hippo core kinases MST1 and MST2. EMBO Mol Med 2017;9(1):46–60. doi:10.15252/emmm.201506089, PMID:27940445.

[35] Zhang C, Bian M, Chen X, Jin H, Zhao S, Yang X, *et al*. Oroxylin A prevents angiogenesis of LSECs in liver fibrosis via inhibition of YAP/HIF-1α signaling. J Cell Biochem 2018;119(2):2258–2268. doi:10.1002/jcb.26388, PMID:28857294.

[36] Song H, Mak KK, Topol L, Yun K, Hu J, Garrett L, *et al*. Mammalian Mst1 and Mst2 kinases play essential roles in organ size control and tumor suppression. Proc Natl Acad Sci U S A 2010;107(4):1431–1436. doi:10.1073/pnas.0911409107, PMID:20080598.

[37] Zhang N, Bai H, David KK, Dong J, Zheng Y, Cai J, *et al*. The Merlin/NF2 tumor suppressor functions through the YAP oncoprotein to regulate tissue homeostasis in mammals. Dev Cell 2010;19(1):27–38. doi:10.1016/j.devcel.2010.06.015, PMID:20643348.

[38] Zhou D, Conrad C, Xia F, Park JS, Payer B, Yin Y, *et al*. Mst1 and Mst2 maintain hepatocyte quiescence and suppress hepatocellular carcinoma development through inactivation of the Yap1 oncogene. Cancer Cell 2009;16(5):425–438. doi:10.1016/j.ccr.2009.09.026, PMID:19878874.

[39] Lee KP, Lee JH, Kim TS, Kim TH, Park HD, Byun JS, *et al*. The Hippo-Salvador pathway restrains hepatic oval cell proliferation, liver size,

and liver tumorigenesis. Proc Natl Acad Sci U S A 2010;107(18):8248–8253. doi:10.1073/pnas.0912203107, PMID:20404163.
[40] Lu L, Li Y, Kim SM, Bossuyt W, Liu P, Qiu Q, *et al*. Hippo signaling is a potent in vivo growth and tumor suppressor pathway in the mammalian liver. Proc Natl Acad Sci U S A 2010;107(4):1437–1442. doi:10.1073/pnas.0911427107, PMID:20080689.
[41] Qi S, Zhong Z, Zhu Y, Wang Y, Ma M, Wang Y, *et al*. Two Hippo signaling modules orchestrate liver size and tumorigenesis. EMBO J 2023;42(22):e115749. doi:10.15252/embj.2023115749, PMID:37849424.
[42] Xiao H, Jiang N, Zhou B, Liu Q, Du C. TAZ regulates cell proliferation and epithelial-mesenchymal transition of human hepatocellular carcinoma. Cancer Sci 2015;106(2):151–159. doi:10.1111/cas.12587, PMID:25495189.
[43] Zhang T, Zhang J, You X, Liu Q, Du Y, Gao Y, *et al*. Hepatitis B virus X protein modulates oncogene Yes-associated protein by CREB to promote growth of hepatoma cells. Hepatology 2012;56(6):2051–2059. doi:10.1002/hep.25899, PMID:22707013.
[44] Overholtzer M, Zhang J, Smolen GA, Muir B, Li W, Sgroi DC, *et al*. Transforming properties of YAP, a candidate oncogene on the chromosome 11q22 amplicon. Proc Natl Acad Sci U S A 2006;103(33):12405–12410. doi:10.1073/pnas.0605579103, PMID:16894141.
[45] Tao J, Calvisi DF, Ranganathan S, Cigliano A, Zhou L, Singh S, *et al*. Activation of β-catenin and Yap1 in human hepatoblastoma and induction of hepatocarcinogenesis in mice. Gastroenterology 2014;147(3):690–701. doi:10.1053/j.gastro.2014.05.004, PMID:24837480.
[46] Moya IM, Castaldo SA, Van den Mooter L, Soheily S, Sansores-Garcia L, Jacobs J, *et al*. Peritumoral activation of the Hippo pathway effectors YAP and TAZ suppresses liver cancer in mice. Science 2019;366(6468):1029–1034. doi:10.1126/science.aaw9886, PMID:31754005.
[47] DeRan M, Yang J, Shen CH, Peters EC, Fitamant J, Chan P, *et al*. Energy stress regulates hippo-YAP signaling involving AMPK-mediated regulation of angiomotin-like 1 protein. Cell Rep 2014;9(2):495–503. doi:10.1016/j.celrep.2014.09.036, PMID:25373897.
[48] Shao D, Zhai P, Del Re DP, Sciarretta S, Yabuta N, Nojima H, *et al*. A functional interaction between Hippo-YAP signalling and FoxO1 mediates the oxidative stress response. Nat Commun 2014;5:3315. doi:10.1038/ncomms4315, PMID:24525530.
[49] Low BC, Pan CQ, Shivashankar GV, Bershadsky A, Sudol M, Sheetz M. YAP/TAZ as mechanosensors and mechanotransducers in regulating organ size and tumor growth. FEBS Lett 2014;588(16):2663–2670. doi:10.1016/j.febslet.2014.04.012, PMID:24747426.
[50] Ma B, Chen Y, Chen L, Cheng H, Mu C, Li J, *et al*. Hypoxia regulates Hippo signalling through the SIAH2 ubiquitin E3 ligase. Nat Cell Biol 2015;17(1):95–103. doi:10.1038/ncb3073, PMID:25438054.
[51] Wu H, Wei L, Fan F, Ji S, Zhang S, Geng J, *et al*. Integration of Hippo signalling and the unfolded protein response to restrain liver overgrowth and tumorigenesis. Nat Commun 2015;6:6239. doi:10.1038/ncomms7239, PMID:25695629.
[52] Lee YA, Noon LA, Akat KM, Ybanez MD, Lee TF, Berres ML, *et al*. Autophagy is a gatekeeper of hepatic differentiation and carcinogenesis by controlling the degradation of Yap. Nat Commun 2018;9(1):4962. doi:10.1038/s41467-018-07338-z, PMID:30470740.
[53] Yang J, Zhang X, Liu L, Yang X, Qian Q, Du B. Corrigendum to: c-Src promotes the growth and tumorigenesis of hepatocellular carcinoma via the Hippo signaling pathway (Life Sci., volume 264(1), article number 118711). Life Sci 2023;330:121932. doi:10.1016/j.lfs.2023.121932, PMID:37586903.
[54] Kwon A, Lee NY, Yu JH, Choi MG, Park J, Koo JH. Mitochondrial stress activates YAP/TAZ through RhoA oxidation to promote liver injury. Cell Death Dis 2024;15(1):51. doi:10.1038/s41419-024-06448-5, PMID:38225223.
[55] Weiler SME, Bissinger M, Rose F, von Bubnoff F, Lutz T, Ori A, *et al*. SEPTIN10-mediated crosstalk between cytoskeletal networks controls mechanotransduction and oncogenic YAP/TAZ signaling. Cancer Lett 2024;584:216637. doi:10.1016/j.canlet.2024.216637, PMID:38242197.
[56] Azzolin L, Zanconato F, Bresolin S, Forcato M, Basso G, Bicciato S, *et al*. Role of TAZ as mediator of Wnt signaling. Cell 2012;151(7):1443–1456. doi:10.1016/j.cell.2012.11.027, PMID:23245942.
[57] Attisano L, Wrana JL. Signal integration in TGF-β, WNT, and Hippo pathways. F1000Prime Rep 2013;5:17. doi:10.12703/P5-17, PMID:23755364.
[58] Kim W, Khan SK, Gvozdenovic-Jeremic J, Kim Y, Dahlman J, Kim H, *et al*. Hippo signaling interactions with Wnt/β-catenin and Notch signaling repress liver tumorigenesis. J Clin Invest 2017;127(1):137–152. doi:10.1172/JCI88486, PMID:27869648.
[59] Jeong SH, Kim HB, Kim MC, Lee JM, Lee JH, Kim JH, *et al*. Hippo-mediated suppression of IRS2/AKT signaling prevents hepatic steatosis and liver cancer. J Clin Invest 2018;128(3):1010–1025. doi:10.1172/JCI95802, PMID:29400692.
[60] Beyer TA, Weiss A, Khomchuk Y, Huang K, Ogunjimi AA, Varelas X, *et al*. Switch enhancers interpret TGF-β and Hippo signaling to control cell fate in human embryonic stem cells. Cell Rep 2013;5(6):1611–1624. doi:10.1016/j.celrep.2013.11.021, PMID:24332857.
[61] Hagenbeek TJ, Webster JD, Kljavin NM, Chang MT, Pham T, Lee HJ, *et al*. The Hippo pathway effector TAZ induces TEAD-dependent liver inflammation and tumors. Sci Signal 2018;11(547):eaaj1757. doi:10.1126/scisignal.aaj1757, PMID:30206136.
[62] Ota M, Sasaki H. Mammalian Tead proteins regulate cell proliferation and contact inhibition as transcriptional mediators of Hippo signaling. Development 2008;135(24):4059–4069. doi:10.1242/dev.027151, PMID:19004856.
[63] Varelas X, Samavarchi-Tehrani P, Narimatsu M, Weiss A, Cockburn K, Larsen BG, *et al*. The Crumbs complex couples cell density sensing to Hippo-dependent control of the TGF-β-SMAD pathway. Dev Cell 2010;19(6):831–844. doi:10.1016/j.devcel.2010.11.012, PMID:21145499.
[64] Varelas X, Sakuma R, Samavarchi-Tehrani P, Peerani R, Rao BM, Dembowy J, *et al*. TAZ controls Smad nucleocytoplasmic shuttling and regulates human embryonic stem-cell self-renewal. Nat Cell Biol 2008;10(7):837–848. doi:10.1038/ncb1748, PMID:18568018.
[65] Labibi B, Bashkurov M, Wrana JL, Attisano L. Modeling the Control of TGF-β/Smad Nuclear Accumulation by the Hippo Pathway Effectors, Taz/Yap. iScience 2020;23(8):101416. doi:10.1016/j.isci.2020.101416, PMID:32798968.
[66] Hiemer SE, Szymaniak AD, Varelas X. The transcriptional regulators TAZ and YAP direct transforming growth factor β-induced tumorigenic phenotypes in breast cancer cells. J Biol Chem 2014;289(19):13461–13474. doi:10.1074/jbc.M113.529115, PMID:24648515.
[67] Oh SH, Swiderska-Syn M, Jewell ML, Premont RT, Diehl AM. Liver regeneration requires Yap1-TGFβ-dependent epithelial-mesenchymal transition in hepatocytes. J Hepatol 2018;69(2):359–367. doi:10.1016/j.jhep.2018.05.008, PMID:29758331.
[68] Malfettone A, Soukupova J, Bertran E, Crosas-Molist E, Lastra R, Fernando J, *et al*. Transforming growth factor-β-induced plasticity causes a migratory stemness phenotype in hepatocellular carcinoma. Cancer Lett 2017;392:39–50. doi:10.1016/j.canlet.2017.01.037, PMID:28161507.
[69] Miyazono K, Katsuno Y, Koinuma D, Ehata S, Morikawa M. Intracellular and extracellular TGF-β signaling in cancer: some recent topics. Front Med 2018;12(4):387–411. doi:10.1007/s11684-018-0646-8, PMID:30043220.
[70] Briones-Orta MA, Delgado-Coello B, Gutiérrez-Vidal R, Sosa-Garrocho M, Macías-Silva M, Mas-Oliva J. Quantitative Expression of Key Cancer Markers in the AS-30D Hepatocarcinoma Model. Front Oncol 2021;11:670292. doi:10.3389/fonc.2021.670292, PMID:34737944.
[71] Xie D, Cui J, Xia T, Jia Z, Wang L, Wei W, *et al*. Hippo transducer TAZ promotes epithelial mesenchymal transition and supports pancreatic cancer progression. Oncotarget 2015;6(34):35949–35963. doi:10.18632/oncotarget.5772, PMID:26416426.
[72] Bhat KP, Salazar KL, Balasubramaniyan V, Wani K, Heathcock L, Hollingsworth F, *et al*. The transcriptional coactivator TAZ regulates mesenchymal differentiation in malignant glioma. Genes Dev 2011;25(24):2594–2609. doi:10.1101/gad.176800.111, PMID:22190458.
[73] Lei QY, Zhang H, Zhao B, Zha ZY, Bai F, Pei XH, *et al*. TAZ promotes cell proliferation and epithelial-mesenchymal transition and is inhibited by the hippo pathway. Mol Cell Biol 2008;28(7):2426–2436.

doi:10.1128/MCB.01874-07, PMID:18227151.
[74] Cordenonsi M, Zanconato F, Azzolin L, Forcato M, Rosato A, Frasson C, *et al*. The Hippo transducer TAZ confers cancer stem cell-related traits on breast cancer cells. Cell 2011;147(4):759–772. doi:10.1016/j.cell.2011.09.048, PMID:22078877.
[75] Hong JH, Hwang ES, McManus MT, Amsterdam A, Tian Y, Kalmukova R, *et al*. TAZ, a transcriptional modulator of mesenchymal stem cell differentiation. Science 2005;309(5737):1074–1078. doi:10.1126/science.1110955, PMID:16099986.
[76] Lin XL, Liu M, Liu Y, Hu H, Pan Y, Zou W, *et al*. Transforming growth factor β1 promotes migration and invasion in HepG2 cells: Epithelial-to-mesenchymal transition via JAK/STAT3 signaling. Int J Mol Med 2018;41(1):129–136. doi:10.3892/ijmm.2017.3228, PMID:29115395.
[77] Yu H, He J, Su G, Wang Y, Fang F, Yang W, *et al*. Fluid shear stress activates YAP to promote epithelial-mesenchymal transition in hepatocellular carcinoma. Mol Oncol 2021;15(11):3164–3183. doi:10.1002/1878-0261.13061, PMID:34260811.
[78] Miranda MZ, Bialik JF, Speight P, Dan Q, Yeung T, Szászi K, *et al*. TGF-β1 regulates the expression and transcriptional activity of TAZ protein via a Smad3-independent, myocardin-related transcription factor-mediated mechanism. J Biol Chem 2017;292(36):14902–14920. doi:10.1074/jbc.M117.780502, PMID:28739802.
[79] Geng J, Yu S, Zhao H, Sun X, Li X, Wang P, *et al*. The transcriptional coactivator TAZ regulates reciprocal differentiation of T(H)17 cells and T(reg) cells. Nat Immunol 2017;18(7):800–812. doi:10.1038/ni.3748, PMID:28504697.
[80] Cohen SB. Our changing environment: an opportunity for physicians. J Med Assoc Ga 1994;83(5):279–280. doi:10.1016/j.heliyon.2023.e21519, PMID:8027697.
[81] Hou Y, Lan C, Kong Y, Zhu C, Peng W, Huang Z, *et al*. Genetic ablation of TAZ induces HepG2 liver cancer cell apoptosis through activating the CaMKII/MIEF1 signaling pathway. Onco Targets Ther 2019;12:1765–1779. doi:10.2147/OTT.S196142, PMID:30881030.
[82] Lee DH, Park JO, Kim TS, Kim SK, Kim TH, Kim MC, *et al*. LATS-YAP/TAZ controls lineage specification by regulating TGFβ signaling and Hnf4α expression during liver development. Nat Commun 2016;7:11961. doi:10.1038/ncomms11961, PMID:27358050.
[83] Yimlamai D, Christodoulou C, Galli GG, Yanger K, Pepe-Mooney B, Gurung B, *et al*. Hippo pathway activity influences liver cell fate. Cell 2014;157(6):1324–1338. doi:10.1016/j.cell.2014.03.060, PMID:24906150.
[84] Noce V, Battistelli C, Cozzolino AM, Consalvi V, Cicchini C, Strippoli R, *et al*. YAP integrates the regulatory Snail/HNF4α circuitry controlling epithelial/hepatocyte differentiation. Cell Death Dis 2019;10(10):768. doi:10.1038/s41419-019-2000-8, PMID:31601778.
[85] Urushima H, Yuasa H, Matsubara T, Kuroda N, Hara Y, Inoue K, *et al*. Activation of Hepatic Stellate Cells Requires Dissociation of E-Cadherin-Containing Adherens Junctions with Hepatocytes. Am J Pathol 2021;191(3):438–453. doi:10.1016/j.ajpath.2020.12.007, PMID:33345995.
[86] Perumal N, Perumal M, Halagowder D, Sivasithamparam N. Morin attenuates diethylnitrosamine-induced rat liver fibrosis and hepatic stellate cell activation by co-ordinated regulation of Hippo/Yap and TGF-β1/Smad signaling. Biochimie 2017;140:10–19. doi:10.1016/j.biochi.2017.05.017, PMID:28552397.
[87] Kim J, Kang W, Kang SH, Park SH, Kim JY, Yang S, *et al*. Proline-rich tyrosine kinase 2 mediates transforming growth factor-beta-induced hepatic stellate cell activation and liver fibrosis. Sci Rep 2020;10(1):21018. doi:10.1038/s41598-020-78056-0, PMID:33273492.
[88] Zhong X, Huang M, Kim HG, Zhang Y, Chowdhury K, Cai W, *et al*. SIRT6 Protects Against Liver Fibrosis by Deacetylation and Suppression of SMAD3 in Hepatic Stellate Cells. Cell Mol Gastroenterol Hepatol 2020;10(2):341–364. doi:10.1016/j.jcmgh.2020.04.005, PMID:32305562.
[89] Zhang J, Li Y, Liu Q, Huang Y, Li R, Wu T, *et al*. Sirt6 Alleviated Liver Fibrosis by Deacetylating Conserved Lysine 54 on Smad2 in Hepatic Stellate Cells. Hepatology 2021;73(3):1140–1157. doi:10.1002/hep.31418, PMID:32535965.
[90] Chowdhury K, Huang M, Kim HG, Dong XC. Sirtuin 6 protects against hepatic fibrogenesis by suppressing the YAP and TAZ function. FASEB J 2022;36(10):e22529. doi:10.1096/fj.202200522R, PMID:36036554.
[91] Nishio M, Sugimachi K, Goto H, Wang J, Morikawa T, Miyachi Y, *et al*. Dysregulated YAP1/TAZ and TGF-β signaling mediate hepatocarcinogenesis in Mob1a/1b-deficient mice. Proc Natl Acad Sci U S A 2016;113(1):E71–E80. doi:10.1073/pnas.1517188113, PMID:26699479.
[92] Sugimachi K, Nishio M, Aishima S, Kuroda Y, Iguchi T, Komatsu H, *et al*. Altered Expression of Hippo Signaling Pathway Molecules in Intrahepatic Cholangiocarcinoma. Oncology 2017;93(1):67–74. doi:10.1159/000463390, PMID:28448997.
[93] Luo W, Li Y, Zeng Y, Li Y, Cheng M, Zhang C, *et al*. Tea domain transcription factor TEAD4 mitigates TGF-β signaling and hepatocellular carcinoma progression independently of YAP. J Mol Cell Biol 2023;15(2):mjad010. doi:10.1093/jmcb/mjad010, PMID:36806855.