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Review Article

The Hippo pathway: an emerging role in urologic cancers

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Abstract: The Hippo pathway controls several biological processes, including cell growth, differentiation, motility, stemness, cell contact, immune cell maturation, organ size, and tumorigenesis. The Hippo pathway core kinases MST1/2 and LATS1/2 in mammals phosphorylate and inactivate YAP1 signaling. Increasing evidence indicates that loss of MST1/2 and LATS1/2 function is linked to the biology of many cancer types with poorer outcomes, likely due to the activation of oncogenic YAP1/TEAD signaling. Therefore, there is a renewed interest in blocking the YAP1/TEAD functions to prevent cancer growth. This review introduces the Hippo pathway components and examines their role and therapeutic potentials in prostate, kidney, and bladder cancer.

Keywords: Hippo pathway, MST1/STK4, MST1/STK3, LATS1, LATS2, YAP1, signal transduction, cancer biology, urologic cancer

Introduction

The Hippo pathway regulates several biological processes, including cell growth, cell fate, energy stress, organ size control, and tumorigenesis [1, 2]. The serine/threonine protein kinases MST1/2 and LATS1/2 (large tumor suppressor 1 and 2) are the core components of the Hippo pathway in mammals [3]. YAP1 (Yes-associated protein 1) and its paralog WWTR1 (WW domain-containing protein; also known as TAZ) are transcriptional coactivators [4, 5]. YAP1 and WWTR1 are key nuclear effectors of the Hippo pathway. MST1/2 and LATS1/2 phosphorylate and inactivate YAP1 by canonical and non-canonical signaling mechanisms. In canonical signaling, MST1/2 phosphorylates and activates LATS1/2 kinase, which in turn phosphorylates and inactivates YAP1 [6]. In non-canonical signaling, however, MST1/2 and LATS1/2 kinase independently phosphorylate and inactivate YAP1 [7, 8]. The MST1/2 and LATS1/2 induction of phospho-Ser127 attenuates the transcriptional activity of YAP1 through cytoplasmic localization and proteasomal degradation via protein 14-3-3 [9, 10].

The TEA domain (TEAD) transcription factors (TEAD1-4) are the critical mediators of YAP1-dependent gene expression [11]. Genes induced by YAP1/TEAD modulate broad cellular processes, including cell growth, migration, survival, anchorage-independent growth, epithelial-mesenchymal transition (EMT), tissue homeostasis, organ size, oncogenic transformation, and tumorigenesis [12]. Also, YAP1 interacts with specific transcriptional programs and signaling pathways that are central to stem cell maintenance and epithelial commitment, including the Wnt/β-catenin [13, 14], Notch [15] and TGF-β [16] pathways. In addition, YAP1 signaling during development has been shown to maintain progenitor populations by enhancing proliferation and simultaneously inhibiting epithelial differentiation [17, 18]. Furthermore, the inactivation of YAP1 resulted in cell contact inhibition and tissue overgrowth [10]. YAP1 also functions as a potent oncogene, and YAP1 nuclear abundance contributes to tumor promotion, progression, and resistance to chemotherapeutics [19]. The interaction between YAP1 and TEAD is mutual because YAP1 serves as a transcriptional coactivator for TEAD-

dependent gene transcriptions. Therefore, targeting the YAP1-TEAD axis is a promising therapeutic strategy against YAP1-induced oncogenesis.

In addition to MST1/2 and LATS1/2, other protein kinases regulate YAP1 signaling possibly in a context-dependent manner [20, 21]. For example, activation of AMPK (AMP-activated protein kinase) under cellular energy stress, such as glucose starvation, disrupted the YAP1-TEAD interaction by directly phosphorylating YAP1 on Ser94, a residue essential for interaction with TEAD [20]. These molecular events inactivated YAP1 and suppressed the growth of LATS-null cells [20]. This study establishes a molecular and functional link between AMPK and the Hippo-YAP1 pathway during cellular energy stress. Moreover, a study by Sorrentino et al. demonstrated that metabolic cues regulate YAP1/TAZ [22]. Mevalonic acid induced by the SREBP transcription factor promotes YAP1 nuclear localization by activating Rho GTPases, which attenuated phospho-Ser127 on YAP1, likely by inhibiting LATS1/2 kinase [23]. Statins, a potent HMG-CoA reductase inhibitor, a rate-limiting enzyme in the SREBP-mevalonate pathway, attenuates YAP1 nuclear localization and transcriptional responses. Mevalonate-YAP1/TAZ axis is required for proliferation and self-renewal of breast cancer cells [22]. In addition, serum-drived S1P and LPA activated YAP1 by inhibiting Hippo/MST signaling, and thus, S1P and LPA might modulate cell proliferation and tumorigenesis by activating YAP1 [24].

Hippo pathway core components

Mammalian STE20-like kinase 1 and 2 (MST1/2, encoded by STK4 and STK3 genes, respectively) are serine-threonine protein kinases [25]. Structurally, MST1/2 has an N-terminal catalytic domain and a C-terminal regulatory domain with SARAH coiled-coiled protein dimerization and inhibitory sites [26]. MST1/2 are phosphoproteins, and phospho-modifications are important for their activation and functions [27]. Autophosphorylation of MST1 on Thr183 (Thr180 in MST2) regulates the MST1/2 kinase activity and apoptosis [28]. Conversely, phosphorylation of MST1 on Thr120 (Thr117 in MST2) by AKT protein kinase could attenuate MST1 activity [29]. In addition, phosphorylation of MST1 at Tyr433 by a non-receptor tyrosine protein kinase c-Abl resulted in MST1 stabilization and activation, leading to neuronal cell death [30]. Similarly, c-Abl was demonstrated to phosphorylate MST2 at Tyr81 and resulted in MST2 activation and neuronal cell death [31]. Nevertheless, phosphorylation of MST1 at Tyr433 by the FGFR4 tyrosine kinase resulted in the inactivation of MST1/2 in T47D and MDA-MB-231 breast cancer cells [32]. In that study, the author showed that the knockdown of FGFR4 promoted MST1 nuclear localization, N-terminal cleavage, and autophosphorylation, which accompanied augmented cell death. In addition, phosphorylation and dimerization was shown to modulate nucleocytoplasmic shuttling of MST1/2 [33]. These observations suggest that MST1/2 can be regulated by a context-dependent manner.

Hippo pathway in development and cancer

Hippo signaling is critical for tissue development and tumorigenesis [34, 35]. Loss of function of the Drosophila MST ortholog, hippo (hpo) caused tissue overgrowth, defects in eye development, and cell enlargement [36, 37]. Similarly, silencing of the hippo-like Cst-1 gene in *C. elegans* reduced life span [38]. Likewise, MST1 or MST2 single gene knockout did not show apparent developmental defects; however, MST1/2 double-knockout mice exhibited early embryonic lethality due to excessive cell death in embryo, developmental defects in placenta, impaired yolk sac/embryo vascular patterning, and primitive hematopoiesis [39].

Evidence based on experimental and clinical studies indicates that loss of MST1/2 signaling results in cancer development [40-42]. Targeted deletion of MST1/2 alleles in the hepatocytes resulted in liver enlargement and eventually caused liver tumors in mice [43]. Similarly, deletion of Sav1 in hepatocytes resulted in hepatic tumors in mice [43]. Transcriptional profiling of both MST1/2 and Sav1 deficient liver tissues revealed a network of genes involved in immune and inflammatory responses [43]. Likewise, deletion of the MST1/2 scaffold protein WW45 (Salvador in Drosophila) in the mouse liver increased liver size and resulted in hepatomas [44]. In addition, deletion of the Nf2/Merlin, an upstream activator of MST1/2, resulted in liver tumors and progressive expansion of progenitor cells in developing

or adult livers without affecting differentiated hepatocytes [45]. MST1/2, WW45, Sav, and Nf2 double mutant liver tissues showed a substantial increase in hepatic progenitor cells or adult facultative stem cells (a.k.a. oval cells, which are commonly associated with liver injury and tumor formation). MST1/2 restrained intestinal stem cell proliferation and colonic tumorigenesis by inhibiting YAP1 nuclear accumulation [46]. A recent study demonstrated that MST1/2 kinases suppressed Ras driven non-small cell lung cancer in the transgenic mouse model [47]. Activation of YAP1 expanded undifferentiated progenitor cells and increased liver size more than 4-fold [48]. These observations are physiologically relevant because the loss of MST1/2 expression have been suggested in head and neck squamous cell carcinoma [49], soft tissue sarcoma [50], glioblastoma [51], and colorectal cancers [52], along with a poorer prognosis.

Hippo pathway in prostate cancer

Metastatic prostate cancer (PC) is a leading cause of cancer deaths among men worldwide. Dysregulated androgen receptor (AR) signaling is central to PC development, progression, metastasis, and relapse. The gene amplification [53], mutations [54], oncogenic growth factor signaling [55], and altered expression of the AR co-regulatory proteins [56] have been shown to dysregulate AR signaling, even in the presence of suboptimal levels of androgens [56, 57]. Therefore, antiandrogen therapy is standard care for patients with advanced PC. This treatment strategy has significant clinical benefits, but it is temporary because the metastatic castration-resistant prostate cancer (CRPC) invariably evolves, even in the presence of second-generation AR inhibitors such as enzalutamide [58, 59]. Despite recent advances [60-69], the molecular mechanisms contributing to CRPC are largely unknown.

Increasing lines of evidence have indicated that loss of MST1/2 functions plays an important role in PC biology [42, 70]. Structural modifications such as by phosphorylation, altered-subcellular localization, and reduced expression by promoter methylation could cause loss of MST1/2 functions [3, 70, 71]. MST1 was initially identified from the AKT protein complexes that were isolated from lipid rafts of LNCaP cells using proteomic approaches [72]. Lipid raft is a cholesterol-rich membrane microdomain and harbors important signals for cell survival [73, 74]. MST1 biochemically interacted with and antagonized AKT signaling in ex vivo and in vivo conditions. In addition, MST1 protein expression was reduced during prostate cancer progression, which coincided with increases in AKT activity. Also, STK4/MST1 functions as a potent negative regulator of AR signaling and suppressor of PC ex vivo and in vivo [75]. These findings are the first to demonstrate that MST1 is a potent inhibitor of AKT and AR oncogenic signaling in PC, supporting the relevance of the Hippo pathway in PC progression. Besides, LATS2 could act as a corepressor by blocking AR protein nuclear-cytoplasmic interactions [76]. The recent studies on genomic and proteomics analyses on PC cells models and clinical samples have shown additional evidence for crucial cellular events related to the aggressiveness of PC, including DNA repair, epigenetic alteration, cell cycle, and translational regulation [77-79].

A growing body of research has indicated that YAP1 activation or amplification is linked to the biology of many cancers with poor prognosis, including PC. YAP1 was demonstrated to transform prostate epithelial cells and promote cell migration, cell invasion, and androgen-independent cell growth, which most likely activated AR and ribosomal S6 kinase (RSK1) signaling [80]. Similarly, induction of KIBRA, a potent activator of YAP1, was shown to promote PC cell proliferation, migration, and invasion in immortalized and cancerous prostate epithelial cells [81]. This study showed that AR promoted KIBRA overexpression, suggesting the functional connection between YAP1 and AR signaling. Moreover, upregulation of YAP1 in the ERG transgenic mouse prostate epithelium resulted in age-related PC [81]. ERG was shown to transcriptionally regulate YAP1 expression and its transcriptional program, providing a possible mechanism by which ERG cooperates with YAP1 to promote PC in mice. Evidence suggests that YAP1 expression was heterogeneous in PC and increased YAP1 expression correlated with PC metastasis to the surrounding tissues [82]. Another study demonstrated that expression of YAP1 increased high-grade PC as opposed to low-grade PC, although neuroendocrine prostate tumors showed reduced

YAP1 expression [82, 83]. Altogether, these studies emphasize the critical role of YAP1 signaling in PC biology.

In addition, an elegant study by Kuser-Abali et al. demonstrated that the interaction of YAP1 with AR may contribute to CRPC [84]. A key finding from this study was that YAP1 and AR interacted with each other without androgen exposure in the CRPC cell model compared to its castration-sensitive PC cell counterpart. This study also showed that genetic silencing of MST1, a potent YAP1 inhibitor, enhanced androgen independent YAP1 and AR interactions. Truncated AR variants, lacking the ligand binding domain, are critical for driving metastatic CRPC [58, 85, 86]. YAP1 interacted with N-terminal domain of AR, providing a possible mechanism of action by which YAP1 mediates development of CRPC cell phenotype in collaboration with AR. A recent study from the same group showed that androgen exposure promoted YAP1 nuclear localization that also occurred in an AR-dependent manner because disruption of AR activity by pharmacologic and genetic approaches reduced the levels of YAP1 protein and nuclear localization [87]. Mechanistically, androgen suppressed the inhibitory phospho-Ser127 on YAP1, possibly activating protein phosphatases and inhibiting MST1 signaling to exert its effect on YAP1. The link between YAP1 and AR is physiologically relevant because the analysis of TCGA (The Cancer Genome Atlas) PC data sets showed that the expression of YAP1 and AR at the transcript levels positively correlate in a subset of PC tissues [87-89]. In addition, a comprehensive analysis of YAP1 protein expression in more than 17,000 prostate cancer specimens showed that YAP1 overexpression is associated with advanced tumor stage, Gleason grade, positive nodal stage, and early biochemical occurrence [90]. Furthermore, enhanced YAP1 immunoreactivity significantly associated with TMPRSS2:ERG fusion, high androgen receptor (AR) expression, high Ki67 labeling index, and PTEN and 8p deletions [90] indicated that high YAP1 expression could be an independent predictor of poorer disease outcomes. Overall, there is a strong connection between YAP1 activation and metastatic CRPC.

Moreover, YAP1 signaling is crucial for maintaining stem cell characteristics. Cancer stem cells are implicated in the etiology of metastatic PC and chemoresistance. A published study suggested that increased YAP1 expression after enzalutamide exposure resulted in overpopulation of cancer stem-like cells [91]. Consistent with this finding, induction of YAP1 promoted cancer stemness and lipid metabolism to mediate the development of enzalutamide-resistant PC [92]. Similarly, a recent study showed that docetaxel exposure elevated the expression of CYR61, YAP1, CD44, CTGF, and ERK in castration-resistant prostate cancer cell lines PC/DX25 and DU/DX50 [93]. Induction of these genes in response to docetaxel could promote migration and invasion abilities of PC/DX25 and DU/DX50 because knockdown of CD44 and YAP1 inhibited observed effects. CYR61, YAP1, CD44, and CTGF are the YAP1 targets, suggesting that higher stem cell populations contribute to resistance to chemotherapeutic agents [93]. Taken together, there is a strong connection between YAP1 activation and the evolution of metastatic PC. Nevertheless, it is unknown whether YAP1 collaborates with AR to contribute to the overpopulation of cancer stem cells in PC in response to cancer therapy.

Furthermore, the impact of tumor microenvironment on cancer progression has gained attention. Cancer-associated fibroblasts (CAFs) are vital components of the tumor microenvironment. Tumor-promoting factors produced by CAFs play important roles in cancer progression and metastasis [94]. A recent study by Shen et al. showed that YAP1 in complex with the TEAD1 transcription factor, a key mediator of YAP1 transcriptional activity, promotes the conversion of normal fibroblasts to CAFs [95]. Mechanistically, the YAP1 and TEAD complex promote CAFs by increasing the expression of SRC, a non-receptor tyrosine kinase, in fibroblasts [95]. The GREM2 (Gremlin 2), a bone morphogenic protein antagonist is considered as another viable target in PC. Shan et al. reported that the elevated miR-423-5p in exosomes secreted by CAFs could lead to taxane resistance targeting GREM2 via the TGF-β pathway [96]. These observations further emphasize the significance of YAP1 signaling in PC progression. Thus, the Hippo/MST1-YAP1-AR axis is a viable cancer drug target to reduce deaths from PC.

Hippo pathway in kidney cancer

Renal cell carcinoma (RCC), which is derived from renal tubular epithelial cells, accounts for

up to 85 percent of all renal malignancies [97]. Over the past 20 years, however, research has revealed that kidney cancer is not a single disease but consisting of multiple dissimilar types of cancer. RCC includes a set of heterogeneous malignancies of the kidney. RCC is one of the most well-known types of cancer, exhibiting 83-88% of human cancer metastasis [98, 99]. However, the kidney cancer types not classified as RCC are graded as non-clear cell RCC (nccRCC). Clear cell RCC (ccRCC), papillary RCC (pRCC), and chromophobe RCC (chRCC) are the main subtypes of kidney cancer with ≥5% occurrence [100]. The other subtypes of kidney cancer are exceptionally uncommon (each with ≤1% occurrence) [101]. In the United States, kidney cancer affects thousands of people each year [102]. However, there is no effective therapy for patients with advanced RCC due to the poorly understood disease mechanisms [102]. Here, we discuss the role of the Hippo-YAP1 in ccRCC, the deadliest form of kidney cancer.

Emerging evidence suggests that dysregulation of Hippo-YAP1 signaling plays a significant role in the etiology of aggressive kidney cancer [103]. A study by Godlewski et al. revealed that YAP1 protein is accumulated in the nuclei of ccRCC cells, even though normal kidney cells primarily express the cytoplasmic YAP1 protein [104]. Likewise, nuclear YAP1 is dramatically higher in ccRCC than nuclear YAP1 in the proximal, unaltered kidney cortexes, as assessed by immunohistochemistry [105]. The upregulation of YAP1 in ccRCC patients showed poorer clinical outcomes [106]. The higher YAP1 protein expression is associated with the clinical stage and pathomorphological features, such as higher TNM and Fuhrman's stages [105]. It appears that nuclear YAP1 has an oncogenic role in ccRCC cells, promoting cell proliferation and survival [105].

Nevertheless, another study demonstrated that cytoplasmic YAP1 correlated with poor prognosis and a high death hazard ratio in the subset of ccRCC patients [107], suggesting that YAP1 retained in the cytoplasm could interact with other signaling pathways to stimulate ccRCC cell proliferation and progression [108]. The proto-oncogene KRAS (Kirsten rat sarcoma virus) acts as a potent oncogene once mutated [109-112]. KRAS is a cytoplasmic and membrane associated protein and a part of the RAS/MAPK pathway [113]. Genetic and biochemical studies suggested that YAP1 and KRAS functionally intersect. For example, YAP1 and KRAS cooperate to regulate the expression of the E2F transcription factor, a key cell cycle regulator [114]. In KRAS-dependent cancer cells, YAP1 functionally counteracted the absence of KRAS oncogenic signaling [115], although the mechanism remains elusive. Other studies showed that cytoplasmic YAP1 correlated with an increase in keratin 19 expression in hepatocellular carcinoma and cholangiocarcinoma. The upregulation of keratin 19 is linked to poor prognosis and cancer progression in patient subsets [116], suggesting the functional interaction between YAP1 and keratin 19 in the development of aggressive cancer. In addition, cytoplasmic YAP1 correlated with histological grade, cancer relapse, and metastasis in uterine cervix squamous cell carcinoma [117].

Moreover, the interaction of YAP1 with the GLI family zinc finger 2 (GLI2) transcription factor promotes the expression of vascular endothelial growth factor A (VEGFA) and angiogenesis in RCC cells [118]. In that study, the author showed that the silencing of YAP1 by RNAi suppressed the angiogenic ability of 786-O kidney cancer cells [118]. Also, the silencing of YAP1 reduced the tube formation and recruitment of human umbilical vein endothelial cells (HUVEC) [119]. The knockdown of GLI2 dramatically reduced YAP1 and VEGFA expression, HUVECs recruitment, and tube formation [118]. GLI2 was demonstrated to promote YAP1 expression, which in turn stimulated the expression of VEGFA in RCC cells [120], implicating that YAP1 is a potential therapeutic target to fight against invasive RCC. In addition, the SRC-JNK (Jun *N*-terminal kinase)-LIMD1 (LIM domains- containing 1)-LATS (large tumor suppressor homolog) axis was demonstrated to promote YAP1 expression through SRC in RCC cells [120]. It was also shown that the activation of SFK (SRC family kinase) and FAK (focal adhesion kinase) upregulated YAP1 in different types of tumors [119-122].

The LATS1/2 kinase phosphorylates inactivates nuclear YAP1 through cytoplasm sequestration [3]. The immunoreactivity of LATS1 was detected in the cytoplasm of normal and cancer cells in the patient subset [123]. However,

the expression of LATS1 was absent or weak in 40% of ccRCC patients [124]. One mechanism suggested that the low levels of LATS1 were due to the hypermethylation of the LATS1 promoter region in ccRCC cells [105]. The demethylation of LATS1 promoter in 786-O cell line is intensely correlated with overexpression of the YAP1 protein [106]. The low levels of LATS1/2 protein are correlated with the clinical stage and pathological grade of ccRCC [104]. A subset of ccRCC patients showed a substantial decline of LATS1/2 protein levels, which is consistent with mRNA levels in another set of patients [125]. Furthermore, the TCGA data from 469 ccRCC tumors revealed that YAP1 mRNA was upregulated in 9.6% and downregulated in 4.9% of cases, and LATS1 mRNA was upregulated in 4.5% and downregulated in 10.7% of cases [4]. The low expression of LATS 1/2 demonstrated a poor survival rate in RCC patients likely due to the nuclear YAP1 abundance.

Furthermore, the SH3 Domain Binding Glutamate Rich Protein Like 2 (SH3BGRL2) was identified as one of the novel regulators of the Hippo pathway in ccRCC. SH3BGRL2 acts as a suppressor via interacting with LATS1/2-YAP1-TEAD1 axis in ccRCC [125]. In addition, YAP1 is transcriptionally activating Twist1 expression by binding to TEAD1, which leads to epithelial-mesenchymal transition (EMT) phenotype [125]. Another regulator is the microphthalmia-associated transcription factor (MITF). MITF is an essential helix-loop-helix leucine zipper transcription factor involved in the progression of various malignancies such as melanoma [126]. In ccRCC, MITF contributes to cell proliferation and tumor growth by activating the RhoA/YAP1 signaling pathway [127]. Silencing MITF hindered the translocation of YAP1 from the cytoplasm to the nucleus [127]. Interestingly, the upregulated YAP1 stimulated cell migration and cell invasion, while these effects were reversed upon MITF silencing [128]. In addition, altered expression of microRNAs (miRNAs) are linked to cancer proliferation in various malignant tumors, including RCC [129-132]. The miR-10b is one of the essential factors in renal cancer [133], and has a vital role in ccRCC cell proliferation, migration, and invasion. Studies demonstrated that miR-10b repressed cell migration and cancer metastasis by targeting HOXA3 through the FAK-YAP1 axis in ccRCC [134].

Degalactotigonin (DGT), a plant extract derived from S. nigrum L, can act as a viable therapeutic agent for advanced RCC. The RNA-seq has demonstrated the efficacy of DGT on 786-O cells affecting YAP1 target genes [135] possibly by inducing the expression of LAST1 and SAV1 that negatively regulate YAP1. In addition, DGT diminished the growth of RCC by YAP1 overexpression in vitro and in vivo. Additional studies suggested that DGT could block YAP1 and TEAD1 interaction, YAP1 expression, and their target gene expression [136]. Also, DGT disrupted YAP1 by stimulating LAST1/2, which leads to YAP1 retention in the cytoplasm [137]. Curcumin is another therapeutic agent that potentially targets YAP1 signaling in Renal Cancer. Curcumin is an herbal compound with anti-cancer effects inhibiting carcinogenesis, angiogenesis, and tumor growth in pre-clinical and clinical studies [138]. Studies demonstrated that the treatment of a low concentration of curcumin stimulated YAP1 and p53 expression but did not induce apoptosis. Nevertheless, the combination of low concentration of curcumin and temsirolimus, an mTOR inhibitor, drastically promoted cell death. Also, high doses of curcumin alone induced apoptosis of the Caki-1 and OSRC-2 renal cell lines [139]. Dasatinib is a pharmacological inhibitor of several tyrosine kinases such as Bcr-Abl and the Src kinase family [140]. Dasatinib was shown to trigger the activation of the JNK-LIMD1-LATS axis and resulted in downregulated YAP1 transcriptional activity in RCC cells [141]. Thus, dasatinib is a promising therapeutic option for RCC in which the Hippo-YAP1 pathway plays a significant role; however, this requires additional studies [120].

Hippo pathway in bladder cancer

Bladder cancer is the fourth most common cancer type with a substantial mortality rate in men and is the eighth most common cancer in women worldwide [142]. The heterogeneity of the disease with the variable pathology of its nature presents a challenge to treat it efficiently. Available molecular data have shown that the pathological properties of bladder cancer are difficult to establish. The complexity arises from different histological subtypes of the disease. Furthermore, lack of standardization on staging, grading, and histological analysis makes comparison of pathological and clinical results on bladder tumor difficult, causing variation in interpretation [143]. In 2016, WHO

(World Health Organization) categorized urothelial cancer into high grade and low grade to create a clear histological difference in between tumors. Three non-invasive group of bladder cancers (pTa low-grade tumors, pTa high-grade tumors, and papillary urothelial neoplasm of low malignant potential (PUNLMP) were included in the list. The non-invasive term differentiates the high- and low-grade papillary carcinomas from invasive urothelial carcinomas. Staging is also another challenge to identify and classify bladder cancer pathological subtypes clearly [143]. Urothelium is one of the slowest cycling epithelia that is exposed to many carcinogens. This makes the bladder a high-risk organ for cancer development, progression, and mortality [144]. About 90% of bladder tumors arise from transitional cells of the urothelium, and the rest of them originates from squamous (5%) and glandular (2%) variants. The remainder of the groups include the rare subtypes of bladder tumors [145, 146].

There are several signaling pathways that involve the survival of bladder cancer cells such as the NF-kB, MAPK, mTOR, and JAK-STAT pathways [147]. The NF-κB pathway has been identified to contribute to the upregulation of the survivin gene in bladder cancer. Studies have also shown that upregulation of the survivin gene by NF-κB not only suppresses apoptosis in bladder cancer cell lines in vivo and in vitro, but it also enhances proliferation [148]. YAP1 has been shown to work as an upstream regulator and activator of the MAPK pathway in bladder cancer [149]. However, the YAP1-MAPK pathway is still a novel area of study in bladder cancer. The YAP1 and mTOR proteins are known to regulate each other positively. The crosstalk between these proteins has been shown to accelerate the progression of the disease [150]. The JAK-STAT pathway is the most studied pathway that has various functions in cellular signal transduction. The deregulation in this pathway is associated with tumorigenesis and metastasis in several cancer types, including bladder cancer [151]. The constitutive activation of STAT3 plays a vital role in bladder malignancy [152]. Chen et al. reported an increase in phospho-STAT3 in bladder cancer tissue and bladder cell lines UMUC-3, WH, and 253-J. The inhibition of STAT3 signaling by dominant negative STAT3-Y705F mutant and small molecule STA-21 inhibitor not only suppressed the bladder cell growth, but also induced apoptosis, demonstrated by immunostaining of cleaved caspases 3, 8, and 9 [152]. Studies have shown that RAC3 (Rac family small GTPase 3) is upregulated in bladder cancer cell lines and tissues [153]. The overexpression of RAC3 could enhance invasion, migration, and proliferation in bladder cancer cells through PYCR1 (pyrroline-5-carboxylate reductase 1), a mitochondrial enzyme, given that PYCR1 knockdown reversed the observed effects of RAC3. Silencing of PYCR1 negatively affects the levels of STAT3, phospho-STAT3, c-MYC, JAK2, and phospho-JAK2 proteins. Overall, activation of the JAK/STAT pathway, which is likely mediated by PYCR1 overexpression, has a critical role in the etiology of bladder cancer [153].

Moreover, recent studies have suggested that Hippo pathway has an important role in the progression of bladder cancer [154]. The Hippo pathway in bladder cancer has not been studied thoroughly. The limited reports have shown that dysregulation of Hippo signaling in bladder cancer is correlated with bladder tumor initiation, progression, and metastasis [155]. Findings point to the fact that the tumor suppressor proteins MST1 and LATS1 are downregulated in bladder cancer clinical samples [156, 157]. Saadeldin et al. identified the alteration and mutations in the LATS1 gene in Egyptian patients with bladder cancer. The group showed that the new variants of LATS1 caused the reduction of LATS1 mRNA expression in urinary bladder tissues [158]. RUNX3 (Runt-related transcription factor 3), which is a downstream effector of the Hippo/MST1 pathway, serves as a tumor suppressor in multiple cancers, including bladder cancer [156]. A recent study investigated the effect of RUNX3 inactivation and polymorphism in bladder cancer [159]. The genetic variations in the RUNX3 gene increases the risk of bladder cancer development and progression [159]. In addition, the role of ETV5, a transcription factor of the ETS family, in FGFR3 and Hippo signaling in bladder cancer has been investigated. The ETV5 is a downstream target of mutant FGFR3 and associated with crosstalk between Hippo and FGFR3 pathway. It is also involved in the up-regulation of genes associated with epithelial-mesenchymal transition of invasive cells, followed by the proliferation and growth of bladder cancer cells [160].

Furthermore, overexpression of YAP1 has also been reported in bladder cancer [161]. 4-Hydroxynonenal (HNE), a pro-oxidant agent, downregulated YAP1 expression via redox-dependent mechanism in bladder cancer cells [162]. Similarly, increasing doses of verteporfin, a potent activator of MST1 kinase and inhibitor of YAP1-TEAD interaction, suppressed bladder cancer cell invasion and growth through MST1/Hippo signaling [160]. In previous studies, YAP1 was noted to promote bladder cancer cell progression and migration by interacting with COX2, ANKRD17, and KLF5 [163-165]. YAP1 expression has also been associated with poor prognosis and the advanced stages of bladder cancer [166]. A recent study suggested that YAP1 could be used as prominent biomarker for shortened survival time in patients with urothelial carcinoma of the bladder [149]. This study indicated that silencing of YAP1 changed the migration and proliferating ability of bladder cancer cell lines [149]. YAP1 promotes cell proliferation and is required for the tumorigenesis of bladder cancer, most likely in collaboration with the MAPK/ERK pathway [149]. All these studies suggest that YAP1 is a prominent target for bladder cancer treatment.

Finally, current therapies focus on improving treatment outcomes using rational cocktail regimens and projectile biomarkers. Although cisplatin-based therapy still reigns as the standard approach at the early metastatic settings, novel therapies are now altering previous treatment paradigms. These therapies include the approval of five immune checkpoint inhibitors that include durvalumab, pembrolizumab, avelumab, nivolumab, and atezolizumab in the platinum refractory setting and two immune checkpoint inhibitors in the first line setting for patients who are deemed cisplatin ineligible and harbor tumors with high PD-L1 expression [156, 157, 159, 160, 162, 167-169]. Studies have shown nivolumab to have significant response rates as well as resilient scientific responses in pretreated metastatic urothelial carcinoma patients [157]. The data from this study, which are consistent with data from previous studies in other malignancies, suggest that there is a substantial benefit in using nivolumab for treating metastatic urothelial carcinoma [157]. Atezolizumab also was proven to have a favorable response and endurance with little incidence of clinically significant toxicities when used in untreated cisplatin-ineligible metastatic urothelial carcinoma patients [159]. In fact, this study suggests that atezolizumab could be a prominent agent for cisplatin-ineligible metastatic urothelial carcinoma [159]. The authors observed that atezolizumab is most efficient when treating metastatic urothelial carcinoma patients with high levels of PD-L1 expression. This theory seems to come from underlying biological and genomic factors [156]. A clinical study investigated the survival rates of 542 patients with urothelial cancer receiving pembrolizumab (200 mg/3 weeks) after platinum chemotherapy [170]. Pembrolizumab is a highly selective monoclonal antibody against programmed death 1 (PD-1) and can disrupt the association between PDL-1 and its ligand that can lead to hampering inhibitory signals in T cells [170]. The overall survival rate of patients was significantly increased by the pembrolizumab treatment (approximately 3 months) compared to chemotherapy alone [170]. In recent studies, enfortumab vedotin (EV) and erdafintib have also been approved for patients who are diagnosed with platinum refractory advanced urinary cancer [171, 172]. Therapeutic therapies for bladder cancer will continue to grow as novel therapeutic targets are discovered. Currently, it is unknown whether any of the immune checkpoint inhibitors tested exert their therapeutic efficacy by modulating the Hippo-YAP1 pathway in bladder cancer.

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Disclosure of conflict of interest

None.

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References

[1] Ma S, Meng Z, Chen R and Guan KL. The Hippo pathway: biology and pathophysiology. Annu Rev Biochem 2019; 88: 577-604.

[2] Mo JS, Park HW and Guan KL. The Hippo signaling pathway in stem cell biology and cancer. EMBO Rep 2014; 15: 642-656.

[3] Meng Z, Moroishi T and Guan KL. Mechanisms of Hippo pathway regulation. Genes Dev 2016; 30: 1-17.

[4] Yu FX, Zhao B and Guan KL. Hippo pathway in organ size control, tissue homeostasis, and cancer. Cell 2015; 163: 811-828.

[5] Chen YA, Lu CY, Cheng TY, Pan SH, Chen HF and Chang NS. WW domain-containing proteins YAP and TAZ in the Hippo pathway as key regulators in stemness maintenance, tissue homeostasis, and tumorigenesis. Front Oncol 2019; 9: 60.

[6] Zhou D, Conrad C, Xia F, Park JS, Payer B, Yin Y, Lauwers GY, Thasler W, Lee JT, Avruch J and Bardeesy N. Mst1 and Mst2 maintain hepatocyte quiescence and suppress hepatocellular carcinoma development through inactivation of the Yap1 oncogene. Cancer Cell 2009; 16: 425-438.

[7] Kuser-Abali G, Alptekin A, Lewis M, Garraway IP and Cinar B. YAP1 and AR interactions contribute to the switch from androgen-dependent to castration-resistant growth in prostate cancer. Nat Commun 2015; 6: 8126.

[8] Yu FX, Zhang Y, Park HW, Jewell JL, Chen Q, Deng Y, Pan D, Taylor SS, Lai ZC and Guan KL. Protein kinase A activates the Hippo pathway to modulate cell proliferation and differentiation. Genes Dev 2013; 27: 1223-1232.

[9] Muslin AJ, Tanner JW, Allen PM and Shaw AS. Interaction of 14-3-3 with signaling proteins is mediated by the recognition of phosphoserine. Cell 1996; 84: 889-897.

[10] Zhao B, Wei X, Li W, Udan RS, Yang Q, Kim J, Xie J, Ikenoue T, Yu J, Li L, Zheng P, Ye K, Chinnaiyan A, Halder G, Lai ZC and Guan KL. Inactivation of YAP oncoprotein by the Hippo pathway is involved in cell contact inhibition and tissue growth control. Genes Dev 2007; 21: 2747-2761.

[11] Li Z, Zhao B, Wang P, Chen F, Dong Z, Yang H, Guan KL and Xu Y. Structural insights into the YAP and TEAD complex. Genes Dev 2010; 24: 235-240.

[12] Huang J, Wu S, Barrera J, Matthews K and Pan D. The Hippo signaling pathway coordinately regulates cell proliferation and apoptosis by inactivating Yorkie, the drosophila homolog of YAP. Cell 2005; 122: 421-434.

[13] Rosenbluh J, Nijhawan D, Cox AG, Li X, Neal JT, Schafer EJ, Zack TI, Wang X, Tsherniak A, Schinzel AC, Shao DD, Schumacher SE, Weir BA, Vazquez F, Cowley GS, Root DE, Mesirov JP, Beroukhim R, Kuo CJ, Goessling W and Hahn WC. β-Catenin-driven cancers require a YAP1 transcriptional complex for survival and tumorigenesis. Cell 2012; 151: 1457-1473.

[14] Konsavage WM Jr, Kyler SL, Rennoll SA, Jin G and Yochum GS. Wnt/β-catenin signaling regulates Yes-associated protein (YAP) gene expression in colorectal carcinoma cells. J Biol Chem 2012; 287: 11730-11739.

[15] Li Y, Hibbs MA, Gard AL, Shylo NA and Yun K. Genome-wide analysis of N1ICD/RBPJ targets in vivo reveals direct transcriptional regulation of Wnt, SHH, and hippo pathway effectors by Notch1. Stem Cells 2012; 30: 741-752.

[16] Aragón E, Goerner N, Zaromytidou AI, Xi Q, Escobedo A, Massagué J and Macias MJ. A Smad action turnover switch operated by WW domain readers of a phosphoserine code. Genes Dev 2011; 25: 1275-1288.

[17] Yu FX, Zhao B, Panupinthu N, Jewell JL, Lian I, Wang LH, Zhao J, Yuan H, Tumaneng K, Li H, Fu XD, Mills GB and Guan KL. Regulation of the Hippo-YAP pathway by G-protein-coupled receptor signaling. Cell 2012; 150: 780-791.

[18] Hiemer SE and Varelas X. Stem cell regulation by the Hippo pathway. Biochim Biophys Acta 2013; 1830: 2323-2334.

[19] Thompson BJ. YAP/TAZ: drivers of tumor growth, metastasis, and resistance to therapy. Bioessays 2010; 42: e1900162.

[20] Mo JS, Meng Z, Kim YC, Park HW, Hansen CG, Kim S, Lim DS and Guan KL. Cellular energy stress induces AMPK-mediated regulation of YAP and the Hippo pathway. Nat Cell Biol 2015; 17: 500-510.

[21] Borreguero-Muñoz N, Fletcher GC, Aguilar-Aragon M, Elbediwy A, Vincent-Mistiaen ZI and Thompson BJ. The Hippo pathway integrates PI3K-Akt signals with mechanical and polarity cues to control tissue growth. PLoS Biol 2019; 17: e3000509.

[22] Sorrentino G, Ruggeri N, Specchia V, Cordenonsi M, Mano M, Dupont S, Manfrin A, Ingallina E, Sommaggio R, Piazza S, Rosato A, Piccolo S and Del Sal G. Metabolic control of YAP and TAZ by the mevalonate pathway. Nat Cell Biol 2014; 16: 357-366.

[23] Mo JS, Yu FX, Gong R, Brown JH and Guan KL. Regulation of the Hippo-YAP pathway by protease-activated receptors (PARs). Genes Dev 2012; 26: 2138-2143.

[24] Miller E, Yang J, DeRan M, Wu C, Su AI, Bonamy GM, Liu J, Peters EC and Wu X. Identification of serum-derived sphingosine-1-phosphate as a small molecule regulator of YAP. Chem Biol 2012; 19: 955-962.

[25] Thompson BJ and Sahai E. MST kinases in development and disease. J Cell Biol 2015; 210: 871-882.

[26] Creasy CL, Ambrose DM and Chernoff J. The Ste20-like protein kinase, Mst1, dimerizes and contains an inhibitory domain. J Biol Chem 1996; 271: 21049-21053.
[27] Rawat SJ and Chernoff J. Regulation of mammalian Ste20 (Mst) kinases. Trends Biochem Sci 2015; 40: 149-156.
[28] Glantschnig H, Rodan GA and Reszka AA. Mapping of MST1 kinase sites of phosphorylation. Activation and autophosphorylation. J Biol Chem 2002; 277: 42987-42996.
[29] Collak FK, Yagiz K, Luthringer DJ, Erkaya B and Cinar B. Threonine-120 phosphorylation regulated by phosphoinositide-3-kinase/Akt and mammalian target of rapamycin pathway signaling limits the antitumor activity of mammalian sterile 20-like kinase 1. J Biol Chem 2012; 287: 23698-23709.
[30] Xiao L, Chen D, Hu P, Wu J, Liu W, Zhao Y, Cao M, Fang Y, Bi W, Zheng Z, Ren J, Ji G, Wang Y and Yuan Z. The c-Abl-MST1 signaling pathway mediates oxidative stress-induced neuronal cell death. J Neurosci 2011; 31: 9611-9619.
[31] Liu W, Wu J, Xiao L, Bai Y, Qu A, Zheng Z and Yuan Z. Regulation of neuronal cell death by c-Abl-Hippo/MST2 signaling pathway. PLoS One 2012; 7: e36562.
[32] Turunen SP, von Nandelstadh P, Öhman T, Gucciardo E, Seashore-Ludlow B, Martins B, Rantanen V, Li H, Höpfner K, Östling P, Varjosalo M and Lehti K. FGFR4 phosphorylates MST1 to confer breast cancer cells resistance to MST1/2-dependent apoptosis. Cell Death Differ 2019; 26: 2577-2593.
[33] Lee KK and Yonehara S. Phosphorylation and dimerization regulate nucleocytoplasmic shuttling of mammalian STE20-like kinase (MST). J Biol Chem 2002; 277: 12351-12358.
[34] Zheng Y and Pan D. The Hippo signaling pathway in development and disease. Dev Cell 2019; 50: 264-282.
[35] Park JH, Shin JE and Park HW. The role of hippo pathway in cancer stem cell biology. Mol Cells 2018; 41: 83-92.
[36] Ai X, Wang D, Zhang J and Shen J. Hippo signaling promotes Ets21c-dependent apical cell extrusion in the Drosophila wing disc. Development 2020; 147: dev190124.
[37] Harvey KF, Pfleger CM and Hariharan IK. The drosophila mst ortholog, hippo, restricts growth and cell proliferation and promotes apoptosis. Cell 2003; 114: 457-467.
[38] Lehtinen MK, Yuan Z, Boag PR, Yang Y, Villén J, Becker EB, DiBacco S, de la Iglesia N, Gygi S, Blackwell TK and Bonni A. A conserved MST-FOXO signaling pathway mediates oxidative-stress responses and extends life span. Cell 2006; 125: 987-1001.
[39] Oh S, Lee D, Kim T, Kim TS, Oh HJ, Hwang CY, Kong YY, Kwon KS and Lim DS. Crucial role for Mst1 and Mst2 kinases in early embryonic development of the mouse. Mol Cell Biol 2009; 29: 6309-6320.
[40] Calses PC, Crawford JJ, Lill JR and Dey A. Hippo pathway in cancer: aberrant regulation and therapeutic opportunities. Trends Cancer 2019; 5: 297-307.
[41] Wang Y, Xu X, Maglic D, Dill MT, Mojumdar K, Ng PK, Jeong KJ, Tsang YH, Moreno D, Bhavana VH, Peng X, Ge Z, Chen H, Li J, Chen Z, Zhang H, Han L, Du D, Creighton CJ and Mills GB; Cancer Genome Atlas Research Network, Camargo F and Liang H. Comprehensive molecular characterization of the Hippo signaling pathway in cancer. Cell Rep 2018; 25: 1304-1317, e1305.
[42] Salem O and Hansen CG. The Hippo pathway in prostate cancer. Cells 2019; 8: 370.
[43] Lu L, Li Y, Kim SM, Bossuyt W, Liu P, Qiu Q, Wang Y, Halder G, Finegold MJ, Lee JS and Johnson RL. 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: 1437-1442.
[44] Lee KP, Lee JH, Kim TS, Kim TH, Park HD, Byun JS, Kim MC, Jeong WI, Calvisi DF, Kim JM and Lim DS. The Hippo-Salvador pathway restrains hepatic oval cell proliferation, liver size, and liver tumorigenesis. Proc Natl Acad Sci U S A 2010; 107: 8248-8253.
[45] Benhamouche S, Curto M, Saotome I, Gladden AB, Liu CH, Giovannini M and McClatchey AI. Nf2/Merlin controls progenitor homeostasis and tumorigenesis in the liver. Genes Dev 2010; 24: 1718-1730.
[46] Zhou D, Zhang Y, Wu H, Barry E, Yin Y, Lawrence E, Dawson D, Willis JE, Markowitz SD, Camargo FD and Avruch J. Mst1 and Mst2 protein kinases restrain intestinal stem cell proliferation and colonic tumorigenesis by inhibition of Yes-associated protein (Yap) overabundance. Proc Natl Acad Sci U S A 2011; 108: E1312-1320.
[47] Singh K, Pruski MA, Polireddy K, Jones NC, Chen Q, Yao J, Dar WA, McAllister F, Ju C, Eltzschig HK, Younes M, Moran C, Karmouty-Quintana H, Ying H and Bailey JM. Mst1/2 kinases restrain transformation in a novel transgenic model of Ras driven non-small cell lung cancer. Oncogene 2020; 39: 1152-1164.
[48] Camargo FD, Gokhale S, Johnnidis JB, Fu D, Bell GW, Jaenisch R and Brummelkamp TR. YAP1 increases organ size and expands undifferentiated progenitor cells. Curr Biol 2007; 17: 2054-2060.
[49] Steinmann K, Sandner A, Schagdarsurengin U and Dammann RH. Frequent promoter hypermethylation of tumor-related genes in head and neck squamous cell carcinoma. Oncol Rep 2009; 22: 1519-1526.

[50] Seidel C, Schagdarsurengin U, Blümke K, Würl P, Pfeifer GP, Hauptmann S, Taubert H and Dammann R. Frequent hypermethylation of MST1 and MST2 in soft tissue sarcoma. Mol Carcinog 2007; 46: 865-871.

[51] Qiao M, Wang Y, Xu X, Lu J, Dong Y, Tao W, Stein J, Stein GS, Iglehart JD, Shi Q and Pardee AB. Mst1 is an interacting protein that mediates PHLPPs' induced apoptosis. Mol Cell 2010; 38: 512-523.

[52] Minoo P, Zlobec I, Baker K, Tornillo L, Terracciano L, Jass JR and Lugli A. Prognostic significance of mammalian sterile20-like kinase 1 in colorectal cancer. Mod Pathol 2007; 20: 331-338.

[53] Waltering KK, Urbanucci A and Visakorpi T. Androgen receptor (AR) aberrations in castration-resistant prostate cancer. Mol Cell Endocrinol 2012; 360: 38-43.

[54] Brooke GN and Bevan CL. The role of androgen receptor mutations in prostate cancer progression. Curr Genomics 2009; 10: 18-25.

[55] Mulholland DJ, Kobayashi N, Ruscetti M, Zhi A, Tran LM, Huang J, Gleave M and Wu H. Pten loss and RAS/MAPK activation cooperate to promote EMT and metastasis initiated from prostate cancer stem/progenitor cells. Cancer Res 2012; 72: 1878-1889.

[56] Culig Z and Santer FR. Androgen receptor coactivators in the regulation of cellular events in prostate cancer. World J Urol 2012; 30: 297-302.

[57] Ishizaki F, Nishiyama T, Kawasaki T, Miyashiro Y, Hara N, Takizawa I, Naito M and Takahashi K. Androgen deprivation promotes intratumoral synthesis of dihydrotestosterone from androgen metabolites in prostate cancer. Sci Rep 2013; 3: 1528.

[58] Antonarakis ES, Lu C, Wang H, Luber B, Nakazawa M, Roeser JC, Chen Y, Mohammad TA, Chen Y, Fedor HL, Lotan TL, Zheng Q, De Marzo AM, Isaacs JT, Isaacs WB, Nadal R, Paller CJ, Denmeade SR, Carducci MA, Eisenberger MA and Luo J. AR-V7 and resistance to enzalutamide and abiraterone in prostate cancer. N Engl J Med 2014; 371: 1028-1038.

[59] Cato L, de Tribolet-Hardy J, Lee I, Rottenberg JT, Coleman I, Melchers D, Houtman R, Xiao T, Li W, Uo T, Sun S, Kuznik NC, Göppert B, Ozgun F, van Royen ME, Houtsmuller AB, Vadhi R, Rao PK, Li L, Balk SP, Den RB, Trock BJ, Karnes RJ, Jenkins RB, Klein EA, Davicioni E, Gruhl FJ, Long HW, Liu XS, Cato ACB, Lack NA, Nelson PS, Plymate SR, Groner AC and Brown M. ARv7 represses tumor-suppressor genes in castration-resistant prostate cancer. Cancer Cell 2019; 35: 401-413, e406.

[60] Cai C, Chen S, Ng P, Bubley GJ, Nelson PS, Mostaghel EA, Marck B, Matsumoto AM, Simon NI, Wang H, Chen S and Balk SP. Intratumoral de novo steroid synthesis activates androgen receptor in castration-resistant prostate cancer and is upregulated by treatment with CYP17A1 inhibitors. Cancer Res 2011; 71: 6503-6513.

[61] Cancer Genome Atlas Research Network. The molecular taxonomy of primary prostate cancer. Cell 2015; 163: 1011-1025.

[62] Chang KH, Li R, Kuri B, Lotan Y, Roehrborn CG, Liu J, Vessella R, Nelson PS, Kapur P, Guo X, Mirzaei H, Auchus RJ and Sharifi N. A gain-of-function mutation in DHT synthesis in castration-resistant prostate cancer. Cell 2013; 154: 1074-1084.

[63] Grasso CS, Wu YM, Robinson DR, Cao X, Dhanasekaran SM, Khan AP, Quist MJ, Jing X, Lonigro RJ, Brenner JC, Asangani IA, Ateeq B, Chun SY, Siddiqui J, Sam L, Anstett M, Mehra R, Prensner JR, Palanisamy N, Ryslik GA, Vandin F, Raphael BJ, Kunju LP, Rhodes DR, Pienta KJ, Chinnaiyan AM and Tomlins SA. The mutational landscape of lethal castration-resistant prostate cancer. Nature 2012; 487: 239-243.

[64] Labrecque MP, Coleman IM, Brown LG, True LD, Kollath L, Lakely B, Nguyen HM, Yang YC, da Costa RMG, Kaipainen A, Coleman R, Higano CS, Yu EY, Cheng HH, Mostaghel EA, Montgomery B, Schweizer MT, Hsieh AC, Lin DW, Corey E, Nelson PS and Morrissey C. Molecular profiling stratifies diverse phenotypes of treatment-refractory metastatic castration-resistant prostate cancer. J Clin Invest 2019; 129: 4492-4505.

[65] Robinson D, Van Allen EM, Wu YM, Schultz N, Lonigro RJ, Mosquera JM, Montgomery B, Taplin ME, Pritchard CC, Attard G, Beltran H, Abida W, Bradley RK, Vinson J, Cao X, Vats P, Kunju LP, Hussain M, Feng FY, Tomlins SA, Cooney KA, Smith DC, Brennan C, Siddiqui J, Mehra R, Chen Y, Rathkopf DE, Morris MJ, Solomon SB, Durack JC, Reuter VE, Gopalan A, Gao J, Loda M, Lis RT, Bowden M, Balk SP, Gaviola G, Sougnez C, Gupta M, Yu EY, Mostaghel EA, Cheng HH, Mulcahy H, True LD, Plymate SR, Dvinge H, Ferraldeschi R, Flohr P, Miranda S, Zafeiriou Z, Tunariu N, Mateo J, Perez-Lopez R, Demichelis F, Robinson BD, Schiffman M, Nanus DM, Tagawa ST, Sigaras A, Eng KW, Elemento O, Sboner A, Heath EI, Scher HI, Pienta KJ, Kantoff P, de Bono JS, Rubin MA, Nelson PS, Garraway LA, Sawyers CL and Chinnaiyan AM. Integrative clinical genomics of advanced prostate cancer. Cell 2015; 161: 1215-1228.

[66] From the American Association of Neurological Surgeons (AANS), American Society of Neuroradiology (ASNR), Cardiovascular and Interventional Radiology Society of Europe (CIRSE), Canadian Interventional Radiology Association (CIRA), Congress of Neurological Surgeons (CNS), European Society of Minimally Invasive

Neurological Therapy (ESMINT), European Society of Neuroradiology (ESNR), European Stroke Organization (ESO), Society for Cardiovascular Angiography and Interventions (SCAI), Society of Interventional Radiology (SIR), Society of NeuroInterventional Surgery (SNIS), and World Stroke Organization (WSO), Sacks D, Baxter B, Campbell BCV, Carpenter JS, Cognard C, Dippel D, Eesa M, Fischer U, Hausegger K, Hirsch JA, Shazam Hussain M, Jansen O, Jayaraman MV, Khalessi AA, Kluck BW, Lavine S, Meyers PM, Ramee S, Rüfenacht DA, Schirmer CM and Vorwerk D. Multisociety consensus quality improvement revised consensus statement for endovascular therapy of acute ischemic stroke. Int J Stroke 2018; 13: 612-632.

[67] Sinha A, Huang V, Livingstone J, Wang J, Fox NS, Kurganovs N, Ignatchenko V, Fritsch K, Donmez N, Heisler LE, Shiah YJ, Yao CQ, Alfaro JA, Volik S, Lapuk A, Fraser M, Kron K, Murison A, Lupien M, Sahinalp C, Collins CC, Tetu B, Masoomian M, Berman DM, van der Kwast T, Bristow RG, Kislinger T and Boutros PC. The proteogenomic landscape of curable prostate cancer. Cancer Cell 2019; 35: 414-427, e416.

[68] Xu K, Wu ZJ, Groner AC, He HH, Cai C, Lis RT, Wu X, Stack EC, Loda M, Liu T, Xu H, Cato L, Thornton JE, Gregory RI, Morrissey C, Vessella RL, Montironi R, Magi-Galluzzi C, Kantoff PW, Balk SP, Liu XS and Brown M. EZH2 oncogenic activity in castration-resistant prostate cancer cells is Polycomb-independent. Science 2012; 338: 1465-1469.

[69] You S, Knudsen BS, Erho N, Alshalalfa M, Takhar M, Al-Deen Ashab H, Davicioni E, Karnes RJ, Klein EA, Den RB, Ross AE, Schaeffer EM, Garraway IP, Kim J and Freeman MR. Integrated classification of prostate cancer reveals a novel luminal subtype with poor outcome. Cancer Res 2016; 76: 4948-4958.

[70] Ready D, Yagiz K, Amin P, Yildiz Y, Funari V, Bozdag S and Cinar B. Mapping the STK4/Hippo signaling network in prostate cancer cell. PLoS One 2017; 12: e0184590.

[71] Kuser-Abali G, Alptekin A and Cinar B. Overexpression of MYC and EZH2 cooperates to epigenetically silence MST1 expression. Epigenetics 2014; 9: 634-643.

[72] Cinar B, Fang PK, Lutchman M, Di Vizio D, Adam RM, Pavlova N, Rubin MA, Yelick PC and Freeman MR. The pro-apoptotic kinase Mst1 and its caspase cleavage products are direct inhibitors of Akt1. EMBO J 2007; 26: 4523-4534.

[73] Simons K and Sampaio JL. Membrane organization and lipid rafts. Cold Spring Harb Perspect Biol 2011; 3: a004697.

[74] Freeman MR, Cinar B and Lu ML. Membrane rafts as potential sites of nongenomic hormonal signaling in prostate cancer. Trends Endocrinol Metab 2005; 16: 273-279.

[75] Cinar B, Collak FK, Lopez D, Akgul S, Mukhopadhyay NK, Kilicarslan M, Gioeli DG and Freeman MR. MST1 is a multifunctional caspase-independent inhibitor of androgenic signaling. Cancer Res 2011; 71: 4303-4313.

[76] Powzaniuk M, McElwee-Witmer S, Vogel RL, Hayami T, Rutledge SJ, Chen F, Harada S, Schmidt A, Rodan GA, Freedman LP and Bai C. The LATS2/KPM tumor suppressor is a negative regulator of the androgen receptor. Mol Endocrinol 2004; 18: 2011-2023.

[77] Schiewer MJ and Knudsen KE. DNA damage response in prostate cancer. Cold Spring Harb Perspect Med 2019; 9: a030486.

[78] Liu Y, Horn JL, Banda K, Goodman AZ, Lim Y, Jana S, Arora S, Germanos AA, Wen L, Hardin WR, Yang YC, Coleman IM, Tharakan RG, Cai EY, Uo T, Pillai SPS, Corey E, Morrissey C, Chen Y, Carver BS, Plymate SR, Beronja S, Nelson PS and Hsieh AC. The androgen receptor regulates a druggable translational regulon in advanced prostate cancer. Sci Transl Med 2019; 11: eaaw4993.

[79] Schiewer MJ, Augello MA and Knudsen KE. The AR dependent cell cycle: mechanisms and cancer relevance. Mol Cell Endocrinol 2012; 352: 34-45.

[80] Zhang L, Yang S, Chen X, Stauffer S, Yu F, Lele SM, Fu K, Datta K, Palermo N, Chen Y and Dong J. The hippo pathway effector YAP regulates motility, invasion, and castration-resistant growth of prostate cancer cells. Mol Cell Biol 2015; 35: 1350-1362.

[81] Stauffer S, Chen X, Zhang L, Chen Y and Dong J. KIBRA promotes prostate cancer cell proliferation and motility. FEBS J 2016; 283: 1800-1811.

[82] Collak FK, Demir U, Ozkanli S, Kurum E and Zerk PE. Increased expression of YAP1 in prostate cancer correlates with extraprostatic extension. Cancer Biol Med 2017; 14: 405-413.

[83] Cheng S, Prieto-Dominguez N, Yang S, Connelly ZM, StPierre S, Rushing B, Watkins A, Shi L, Lakey M, Baiamonte LB, Fazili T, Lurie A, Corey E, Shi R, Yeh Y and Yu X. The expression of YAP1 is increased in high-grade prostatic adenocarcinoma but is reduced in neuroendocrine prostate cancer. Prostate Cancer Prostatic Dis 2020; 23: 661-669.

[84] Kuser-Abali G, Alptekin A, Lewis M, Garraway IP and Cinar B. YAP1 and AR interactions contribute to the switch from androgen-dependent to castration-resistant growth in prostate cancer. Nat Commun 2015; 6: 8126.

[85] Tietz KT and Dehm SM. Androgen receptor variants: RNA-based mechanisms and therapeutic targets. Hum Mol Genet 2020; 29: R19-R26.

[86] Vlachostergios PJ, Puca L and Beltran H. Emerging variants of castration-resistant prostate cancer. Curr Oncol Rep 2017; 19: 32.

[87] Cinar B, Al-Mathkour MM, Khan SA and Moreno CS. Androgen attenuates the inactivating phospho-Ser-127 modification of yes-associated protein 1 (YAP1) and promotes YAP1 nuclear abundance and activity. J Biol Chem 2020; 295: 8550-8559.

[88] Gao J, Aksoy BA, Dogrusoz U, Dresdner G, Gross B, Sumer SO, Sun Y, Jacobsen A, Sinha R, Larsson E, Cerami E, Sander C and Schultz N. Integrative analysis of complex cancer genomics and clinical profiles using the cBioPortal. Sci Signal 2013; 6: pl1.

[89] Sanchez-Vega F, Mina M, Armenia J, Chatila WK, Luna A, La KC, Dimitriadoy S, Liu DL, Kantheti HS, Saghafinia S, Chakravarty D, Daian F, Gao Q, Bailey MH, Liang WW, Foltz SM, Shmulevich I, Ding L, Heins Z, Ochoa A, Gross B, Gao J, Zhang H, Kundra R, Kandoth C, Bahceci I, Dervishi L, Dogrusoz U, Zhou W, Shen H, Laird PW, Way GP, Greene CS, Liang H, Xiao Y, Wang C, Iavarone A, Berger AH, Bivona TG, Lazar AJ, Hammer GD, Giordano T, Kwong LN, McArthur G, Huang C, Tward AD, Frederick MJ, McCormick F, Meyerson M; Cancer Genome Atlas Research Network, Van Allen EM, Cherniack AD, Ciriello G, Sander C and Schultz N. Oncogenic signaling pathways in the cancer genome atlas. Cell 2018; 173: 321-337, e310.

[90] Marx A, Schumann A, Höflmayer D, Bady E, Hube-Magg C, Möller K, Tsourlakis MC, Steurer S, Büscheck F, Eichenauer T, Clauditz TS, Graefen M, Simon R, Sauter G, Izbicki JR, Huland H, Heinzer H, Haese A, Schlomm T, Bernreuther C, Lebok P and Polonski A. Up regulation of the Hippo signalling effector YAP1 is linked to early biochemical recurrence in prostate cancers. Sci Rep 2020; 10: 8916.

[91] Jiang N, Ke B, Hjort-Jensen K, Iglesias-Gato D, Wang Z, Chang P, Zhao Y, Niu X, Wu T, Peng B, Jiang M, Li X, Shang Z, Wang Q, Chang C, Flores-Morales A and Niu Y. YAP1 regulates prostate cancer stem cell-like characteristics to promote castration resistant growth. Oncotarget 2017; 8: 115054-115067.

[92] Lee HC, Ou CH, Huang YC, Hou PC, Creighton CJ, Lin YS, Hu CY and Lin SC. YAP1 overexpression contributes to the development of enzalutamide resistance by induction of cancer stemness and lipid metabolism in prostate cancer. Oncogene 2021; 40: 2407-2421.

[93] Lai CJ, Lin CY, Liao WY, Hour TC, Wang HD and Chuu CP. CD44 promotes migration and invasion of docetaxel-resistant prostate cancer cells likely via induction of Hippo-yap signaling. Cells 2019; 8: 295.

[94] Cheteh EH, Augsten M, Rundqvist H, Bianchi J, Sarne V, Egevad L, Bykov VJ, Östman A and Wiman KG. Human cancer-associated fibroblasts enhance glutathione levels and antagonize drug-induced prostate cancer cell death. Cell Death Dis 2017; 8: e2848.

[95] Shen T, Li Y, Zhu S, Yu J, Zhang B, Chen X, Zhang Z, Ma Y, Niu Y and Shang Z. YAP1 plays a key role of the conversion of normal fibroblasts into cancer-associated fibroblasts that contribute to prostate cancer progression. J Exp Clin Cancer Res 2020; 39: 36.

[96] Shan G, Gu J, Zhou D, Li L, Cheng W, Wang Y, Tang T and Wang X. Cancer-associated fibroblast-secreted exosomal miR-423-5p promotes chemotherapy resistance in prostate cancer by targeting GREM2 through the TGF-β signaling pathway. Exp Mol Med 2020; 52: 1809-1822.

[97] Kovacs G, Akhtar M, Beckwith BJ, Bugert P, Cooper CS, Delahunt B, Eble JN, Fleming S, Ljungberg B, Medeiros LJ, Moch H, Reuter VE, Ritz E, Roos G, Schmidt D, Srigley JR, Störkel S, van den Berg E and Zbar B. The Heidelberg classification of renal cell tumours. J Pathol 1997; 183: 131-133.

[98] Vera-Badillo FE, Templeton AJ, Duran I, Ocana A, de Gouveia P, Aneja P, Knox JJ, Tannock IF, Escudier B and Amir E. Systemic therapy for non-clear cell renal cell carcinomas: a systematic review and meta-analysis. Eur Urol 2015; 67: 740-749.

[99] Kroeger N, Xie W, Lee JL, Bjarnason GA, Knox JJ, Mackenzie MJ, Wood L, Srinivas S, Vaishamayan UN, Rha SY, Pal SK, Yuasa T, Donskov F, Agarwal N, Kollmannsberger CK, Tan MH, North SA, Rini BI, Choueiri TK and Heng DY. Metastatic non-clear cell renal cell carcinoma treated with targeted therapy agents: characterization of survival outcome and application of the International mRCC Database Consortium criteria. Cancer 2013; 119: 2999-3006.

[100] Cancer Genome Atlas Research Network. Comprehensive molecular characterization of clear cell renal cell carcinoma. Nature 2013; 499: 43-49.

[101] Moch H, Cubilla AL, Humphrey PA, Reuter VE and Ulbright TM. The 2016 WHO classification of tumours of the urinary system and male genital organs-part A: renal, penile, and testicular tumours. Eur Urol 2016; 70: 93-105.

[102] Linehan WM, Walther MM and Zbar B. The genetic basis of cancer of the kidney. J Urol 2003; 170: 2163-2172.

[103] Han Y. Analysis of the role of the Hippo pathway in cancer. J Transl Med 2019; 17: 116.

[104] Godlewski J, Kiezun J, Krazinski BE, Kozielec Z, Wierzbicki PM and Kmiec Z. The immunoexpression of YAP1 and LATS1 proteins in clear cell renal cell carcinoma: impact on pa-

tients' survival. Biomed Res Int 2018; 2018: 2653623.
[105] Rybarczyk A, Klacz J, Wronska A, Matuszewski M, Kmiec Z and Wierzbicki PM. Overexpression of the YAP1 oncogene in clear cell renal cell carcinoma is associated with poor outcome. Oncol Rep 2017; 38: 427-439.
[106] Chen KH, He J, Wang DL, Cao JJ, Li MC, Zhao XM, Sheng X, Li WB and Liu WJ. Methylation-associated inactivation of LATS1 and its effect on demethylation or overexpression on YAP and cell biological function in human renal cell carcinoma. Int J Oncol 2014; 45: 2511-2521.
[107] Sun Z, Xu R, Li X, Ren W, Ou C, Wang Q, Zhang H, Zhang X, Ma J, Wang H and Li G. Prognostic value of yes-associated protein 1 (YAP1) in various cancers: a meta-analysis. PLoS One 2015; 10: e0135119.
[108] Shreberk-Shaked M and Oren M. New insights into YAP/TAZ nucleo-cytoplasmic shuttling: new cancer therapeutic opportunities? Mol Oncol 2019; 13: 1335-1341.
[109] Tsuchida N, Ryder T and Ohtsubo E. Nucleotide sequence of the oncogene encoding the p21 transforming protein of Kirsten murine sarcoma virus. Science 1982; 217: 937-939.
[110] Chiosea SI, Sherer CK, Jelic T and Dacic S. KRAS mutant allele-specific imbalance in lung adenocarcinoma. Mod Pathol 2011; 24: 1571-1577.
[111] Hartman DJ, Davison JM, Foxwell TJ, Nikiforova MN and Chiosea SI. Mutant allele-specific imbalance modulates prognostic impact of KRAS mutations in colorectal adenocarcinoma and is associated with worse overall survival. Int J Cancer 2012; 131: 1810-1817.
[112] Krasinskas AM, Moser AJ, Saka B, Adsay NV and Chiosea SI. KRAS mutant allele-specific imbalance is associated with worse prognosis in pancreatic cancer and progression to undifferentiated carcinoma of the pancreas. Mod Pathol 2013; 26: 1346-1354.
[113] Welman A, Burger MM and Hagmann J. Structure and function of the C-terminal hypervariable region of K-Ras4B in plasma membrane targetting and transformation. Oncogene 2000; 19: 4582-91.
[114] Sharif GM, Schmidt MO, Yi C, Hu Z, Haddad BR, Glasgow E, Riegel AT and Wellstein A. Cell growth density modulates cancer cell vascular invasion via Hippo pathway activity and CXCR2 signaling. Oncogene 2015; 34: 5879-5889.
[115] Kapoor A, Yao W, Ying H, Hua S, Liewen A, Wang Q, Zhong Y, Wu CJ, Sadanandam A, Hu B, Chang Q, Chu GC, Al-Khalil R, Jiang S, Xia H, Fletcher-Sananikone E, Lim C, Horwitz GI, Viale A, Pettazzoni P, Sanchez N, Wang H, Protopopov A, Zhang J, Heffernan T, Johnson RL, Chin L, Wang YA, Draetta G and DePinho RA. Yap1 activation enables bypass of oncogenic Kras addiction in pancreatic cancer. Cell 2014; 158: 185-19.
[116] Lee K, Lee KB, Jung HY, Yi NJ, Lee KW, Suh KS and Jang JJ. The correlation between poor prognosis and increased yes-associated protein 1 expression in keratin 19 expressing hepatocellular carcinomas and cholangiocarcinomas. BMC Cancer 2017; 17: 441.
[117] Liu T, Liu Y, Gao H, Meng F, Yang S and Lou G. Clinical significance of yes-associated protein overexpression in cervical carcinoma: the differential effects based on histotypes. Int J Gynecol Cancer 2013; 23: 735-742.
[118] Xu S, Zhang H, Chong Y, Guan B and Guo P. YAP promotes VEGFA expression and tumor angiogenesis though Gli2 in human renal cell carcinoma. Arch Med Res 2019; 50: 225-233.
[119] Xiang X, Wang Y, Zhang H, Piao J, Muthusamy S, Wang L, Deng Y, Zhang W, Kuang R, Billadeau DD, Huang S, Lai J, Urrutia R and Kang N. Vasodilator-stimulated phosphoprotein promotes liver metastasis of gastrointestinal cancer by activating a beta1-integrin-FAK-YAP1/TAZ signaling pathway. NPJ Precis Oncol 2018; 2: 2.
[120] Sun J, Wang X, Tang B, Liu H, Zhang M, Wang Y, Ping F, Ding J, Shen A and Geng M. A tightly controlled Src-YAP signaling axis determines therapeutic response to dasatinib in renal cell carcinoma. Theranostics 2018; 8: 3256-3267.
[121] Taniguchi K, Wu LW, Grivennikov SI, de Jong PR, Lian I, Yu FX, Wang K, Ho SB, Boland BS, Chang JT, Sandborn WJ, Hardiman G, Raz E, Maehara Y, Yoshimura A, Zucman-Rossi J, Guan KL and Karin M. A gp130-Src-YAP module links inflammation to epithelial regeneration. Nature 2015; 519: 57-62.
[122] Lamar JM, Xiao Y, Norton E, Jiang ZG, Gerhard GM, Kooner S, Warren JSA and Hynes RO. SRC tyrosine kinase activates the YAP/TAZ axis and thereby drives tumor growth and metastasis. J Biol Chem 2019; 294: 2302-2317.
[123] Zanconato F, Cordenonsi M and Piccolo S. YAP/TAZ at the roots of cancer. Cancer Cell 2016; 29: 783-803.
[124] Visser S and Yang X. LATS tumor suppressor: a new governor of cellular homeostasis. Cell Cycle 2010; 9: 3892-3903.
[125] Yin L, Li W, Xu A, Shi H, Wang K, Yang H, Wang R and Peng B. SH3BGRL2 inhibits growth and metastasis in clear cell renal cell carcinoma via activating hippo/TEAD1-Twist1 pathway. EBioMedicine 2020; 51: 102596.
[126] Widlund HR and Fisher DE. Microphthalamia-associated transcription factor: a critical regulator of pigment cell development and survival. Oncogene 2003; 22: 3035-3041.
[127] Kim N, Kim S, Lee MW, Jeon HJ, Ryu H, Kim JM and Lee HJ. MITF Promotes cell growth, migration and invasion in clear cell renal cell carci-

noma by activating the RhoA/YAP signal pathway. Cancers (Basel) 2021; 13: 2920.
[128] Dupont S, Morsut L, Aragona M, Enzo E, Giulitti S, Cordenonsi M, Zanconato F, Le Digabel J, Forcato M, Bicciato S, Elvassore N and Piccolo S. Role of YAP/TAZ in mechanotransduction. Nature 2011; 474: 179-183.
[129] Han X, Yan S, Weijie Z, Feng W, Liuxing W, Mengquan L and Qingxia F. Critical role of miR-10b in transforming growth factor-beta1-induced epithelial-mesenchymal transition in breast cancer. Cancer Gene Ther 2014; 21: 60-67.
[130] Bloomston M, Frankel WL, Petrocca F, Volinia S, Alder H, Hagan JP, Liu CG, Bhatt D, Taccioli C and Croce CM. MicroRNA expression patterns to differentiate pancreatic adenocarcinoma from normal pancreas and chronic pancreatitis. JAMA 2007; 297: 1901-1908.
[131] Ladeiro Y, Couchy G, Balabaud C, Bioulac-Sage P, Pelletier L, Rebouissou S and Zucman-Rossi J. MicroRNA profiling in hepatocellular tumors is associated with clinical features and oncogene/tumor suppressor gene mutations. Hepatology 2008; 47: 1955-1963.
[132] Xiao H, Li H, Yu G, Xiao W, Hu J, Tang K, Zeng J, He W, Zeng G, Ye Z and Xu H. MicroRNA-10b promotes migration and invasion through KLF4 and HOXD10 in human bladder cancer. Oncol Rep 2014; 31: 1832-1838.
[133] Heinzelmann J, Henning B, Sanjmyatav J, Posorski N, Steiner T, Wunderlich H, Gajda MR and Junker K. Specific miRNA signatures are associated with metastasis and poor prognosis in clear cell renal cell carcinoma. World J Urol 2011; 29: 367-373.
[134] He C, Chen ZY, Li Y, Yang ZQ, Zeng F, Cui Y, He Y, Chen JB and Chen HQ. miR-10b suppresses cell invasion and metastasis through targeting HOXA3 regulated by FAK/YAP signaling pathway in clear-cell renal cell carcinoma. BMC Nephrol 2019; 20: 127.
[135] Wang Y, et al. The natural extract degalactotigonin exerts antitumor effects on renal cell carcinoma cells through repressing YAP. Translational Cancer Research 2020; 9: 7550-7561.
[136] Wong JS, Meliambro K, Ray J and Campbell KN. Hippo signaling in the kidney: the good and the bad. Am J Physiol Renal Physiol 2016; 311: F241-248.
[137] Qu L, Wu Z, Li Y, Xu Z, Liu B, Liu F, Bao Y, Wu D, Liu J, Wang A, Chu X, Sun Y, Chen C, Zhang Z and Wang L. A feed-forward loop between lncARSR and YAP activity promotes expansion of renal tumour-initiating cells. Nat Commun 2016; 7: 12692.
[138] Mansouri K, Rasoulpoor S, Daneshkhah A, Abolfathi S, Salari N, Mohammadi M, Rasoulpoor S and Shabani S. Clinical effects of curcumin in enhancing cancer therapy: a systematic review. BMC Cancer 2020; 20: 791.
[139] Xu S, Yang Z, Fan Y, Guan B, Jia J, Gao Y, Wang K, Wu K, Wang X, Zheng P, He D and Guo P. Curcumin enhances temsirolimus-induced apoptosis in human renal carcinoma cells through upregulation of YAP/p53. Oncol Lett 2016; 12: 4999-5006.
[140] Lombardo LJ, Lee FY, Chen P, Norris D, Barrish JC, Behnia K, Castaneda S, Cornelius LA, Das J, Doweyko AM, Fairchild C, Hunt JT, Inigo I, Johnston K, Kamath A, Kan D, Klei H, Marathe P, Pang S, Peterson R, Pitt S, Schieven GL, Schmidt RJ, Tokarski J, Wen ML, Wityak J and Borzilleri RM. Discovery of N-(2-chloro-6-methyl-phenyl)-2-(6-(4-(2-hydroxyethyl)-piperazin-1-yl)-2-methylpyrimidin-4-ylamino)thiazole-5-carboxamide (BMS-354825), a dual Src/Abl kinase inhibitor with potent antitumor activity in preclinical assays. J Med Chem 2004; 47: 6658-6661.
[141] Shi H, Zhang CJ, Chen GY and Yao SQ. Cell-based proteome profiling of potential dasatinib targets by use of affinity-based probes. J Am Chem Soc 2012; 134: 3001-3014.
[142] Oeyen E, Hoekx L, De Wachter S, Baldewijns M, Ameye F and Mertens I. Bladder cancer diagnosis and follow-up: the current status and possible role of extracellular vesicles. Int J Mol Sci 2019; 20: 821.
[143] Compérat E, Varinot J, Moroch J, Eymerit-Morin C and Brimo F. A practical guide to bladder cancer pathology. Nat Rev Urol 2018; 15: 143-154.
[144] Apodaca G. The uroepithelium: not just a passive barrier. Traffic 2004; 5: 117-128.
[145] Ferlay J, Soerjomataram I, Dikshit R, Eser S, Mathers C, Rebelo M, Parkin DM, Forman D and Bray F. Cancer incidence and mortality worldwide: sources, methods and major patterns in GLOBOCAN 2012. Int J Cancer 2015; 136: E359-E386.
[146] Reuter VE. The pathology of bladder cancer. Urology 2006; 67: 11-17; discussion 17-18.
[147] Abbosh PH, McConkey DJ and Plimack ER. Targeting signaling transduction pathways in bladder cancer. Curr Oncol Rep 2015; 17: 58.
[148] Cui X, Shen D, Kong C, Zhang Z, Zeng Y, Lin X and Liu X. NF-κB suppresses apoptosis and promotes bladder cancer cell proliferation by upregulating survivin expression in vitro and in vivo. Sci Rep 2017; 7: 40723.
[149] Qiu D, Zhu Y and Cong Z. YAP triggers bladder cancer proliferation by affecting the MAPK pathway. Cancer Manag Res 2020; 12: 12205-12214.
[150] Xu M, Gu M, Zhou J, Da J and Wang Z. Interaction of YAP1 and mTOR promotes bladder can-

cer progression. Int J Oncol 2020; 56: 232-242.
[151] Degoricija M, Situm M, Korać J, Miljković A, Matić K, Paradžik M, Marinović Terzić I, Jerončić A, Tomić S and Terzić J. High NF-κB and STAT3 activity in human urothelial carcinoma: a pilot study. World J Urol 2014; 32: 1469-1475.
[152] Chen CL, Cen L, Kohout J, Hutzen B, Chan C, Hsieh FC, Loy A, Huang V, Cheng G and Lin J. Signal transducer and activator of transcription 3 activation is associated with bladder cancer cell growth and survival. Mol Cancer 2008; 7: 78-78.
[153] Cheng C, Song D, Wu Y and Liu B. RAC3 promotes proliferation, migration and invasion via PYCR1/JAK/STAT signaling in bladder cancer. Front Mol Biosci 2020; 7: 218.
[154] Xia J, Zeng M, Zhu H, Chen X, Weng Z and Li S. Emerging role of Hippo signalling pathway in bladder cancer. J Cell Mol Med 2018; 22: 4-15.
[155] Lotan Y, Kamat AM, Porter MP, Robinson VL, Shore N, Jewett M, Schelhammer PF, deVere White R, Quale D and Lee CT; Bladder Cancer Think Tank; Bladder Cancer Advocacy Network; Society of Urologic Oncology. Key concerns about the current state of bladder cancer: a position paper from the Bladder Cancer Think Tank, the Bladder Cancer Advocacy Network, and the Society of Urologic Oncology. Cancer 2005; 115: 4096-4103.
[156] Rosenberg JE, Hoffman-Censits J, Powles T, van der Heijden MS, Balar AV, Necchi A, Dawson N, O'Donnell PH, Balmanoukian A, Loriot Y, Srinivas S, Retz MM, Grivas P, Joseph RW, Galsky MD, Fleming MT, Petrylak DP, Perez-Gracia JL, Burris HA, Castellano D, Canil C, Bellmunt J, Bajorin D, Nickles D, Bourgon R, Frampton GM, Cui N, Mariathasan S, Abidoye O, Fine GD and Dreicer R. Atezolizumab in patients with locally advanced and metastatic urothelial carcinoma who have progressed following treatment with platinum-based chemotherapy: a single-arm, multicentre, phase 2 trial. Lancet 2016; 387: 1909-1920.
[157] Sharma P, Callahan MK, Bono P, Kim J, Spiliopoulou P, Calvo E, Pillai RN, Ott PA, de Braud F, Morse M, Le DT, Jaeger D, Chan E, Harbison C, Lin CS, Tschaika M, Azrilevich A and Rosenberg JE. Nivolumab monotherapy in recurrent metastatic urothelial carcinoma (CheckMate 032): a multicentre, open-label, two-stage, multiarm, phase 1/2 trial. Lancet Oncol 2016; 17: 1590-1598.
[158] Saadeldin MK, Shawer H, Mostafa A, Kassem NM, Amleh A and Siam R. New genetic variants of LATS1 detected in urinary bladder and colon cancer. Front Genet 2015; 5: 425.
[159] Balar AV, Galsky MD, Rosenberg JE, Powles T, Petrylak DP, Bellmunt J, Loriot Y, Necchi A, Hoffman-Censits J, Perez-Gracia JL, Dawson NA, van der Heijden MS, Dreicer R, Srinivas S, Retz MM, Joseph RW, Drakaki A, Vaishampayan UN, Sridhar SS, Quinn DI, Durán I, Shaffer DR, Eigl BJ, Grivas PD, Yu EY, Li S, Kadel EE 3rd, Boyd Z, Bourgon R, Hegde PS, Mariathasan S, Thåström A, Abidoye OO, Fine GD and Bajorin DF; IMvigor210 Study Group. Atezolizumab as first-line treatment in cisplatin-ineligible patients with locally advanced and metastatic urothelial carcinoma: a single-arm, multicentre, phase 2 trial. Lancet 2017; 389: 67-76.
[160] Bellmunt J, de Wit R, Vaughn DJ, Fradet Y, Lee JL, Fong L, Vogelzang NJ, Climent MA, Petrylak DP, Choueiri TK, Necchi A, Gerritsen W, Gurney H, Quinn DI, Culine S, Sternberg CN, Mai Y, Poehlein CH, Perini RF and Bajorin DF; KEYNOTE-045 Investigators. Pembrolizumab as second-line therapy for advanced urothelial carcinoma. N Engl J Med 2017; 376: 1015-1026.
[161] Abugomaa A, Elbadawy M, Yamawaki H, Usui T and Sasaki K. Emerging roles of cancer stem cells in bladder cancer progression, tumorigenesis, and resistance to chemotherapy: a potential therapeutic target for bladder cancer. Cells 2020; 9: 235.
[162] Powles T, O'Donnell PH, Massard C, Arkenau HT, Friedlander TW, Hoimes CJ, Lee JL, Ong M, Sridhar SS, Vogelzang NJ, Fishman MN, Zhang J, Srinivas S, Parikh J, Antal J, Jin X, Gupta AK, Ben Y and Hahn NM. Efficacy and safety of durvalumab in locally advanced or metastatic urothelial carcinoma: updated results from a phase 1/2 open-label study. JAMA Oncol 2017; 3: e172411.
[163] Ooki A, Del Carmen Rodriguez Pena M, Marchionni L, Dinalankara W, Begum A, Hahn NM, VandenBussche CJ, Rasheed ZA, Mao S, Netto GJ, Sidransky D and Hoque MO. YAP1 and COX2 coordinately regulate urothelial cancer stem-like cells. Cancer Res 2018; 78: 168-181.
[164] Dong L, Lin F, Wu W, Huang W and Cai Z. Transcriptional cofactor Mask2 is required for YAP-induced cell growth and migration in bladder cancer cell. J Cancer 2016; 7: 2132-2138.
[165] Gao Y, Shi Q, Xu S, Du C, Liang L, Wu K, Wang K, Wang X, Chang LS, He D and Guo P. Curcumin promotes KLF5 proteasome degradation through downregulating YAP/TAZ in bladder cancer cells. Int J Mol Sci 2014; 15: 15173-15187.
[166] Liu JY, Li YH, Lin HX, Liao YJ, Mai SJ, Liu ZW, Zhang ZL, Jiang LJ, Zhang JX, Kung HF, Zeng YX, Zhou FJ and Xie D. Overexpression of YAP 1 contributes to progressive features and poor prognosis of human urothelial carcinoma of the bladder. BMC Cancer 2013; 13: 349-349.

[167] Robertson AG, Kim J, Al-Ahmadie H, Bellmunt J, Guo G, Cherniack AD, Hinoue T, Laird PW, Hoadley KA, Akbani R, Castro MAA, Gibb EA, Kanchi RS, Gordenin DA, Shukla SA, Sanchez-Vega F, Hansel DE, Czerniak BA, Reuter VE, Su X, de Sa Carvalho B, Chagas VS, Mungall KL, Sadeghi S, Pedamallu CS, Lu Y, Klimczak LJ, Zhang J, Choo C, Ojesina AI, Bullman S, Leraas KM, Lichtenberg TM, Wu CJ, Schultz N, Getz G, Meyerson M, Mills GB, McConkey DJ; TCGA Research Network, Weinstein JN, Kwiatkowski DJ and Lerner SP. Comprehensive molecular characterization of muscle-invasive bladder cancer. Cell 2017; 171: 540-556, e525.

[168] Patel MR, Ellerton J, Infante JR, Agrawal M, Gordon M, Aljumaily R, Britten CD, Dirix L, Lee KW, Taylor M, Schöffski P, Wang D, Ravaud A, Gelb AB, Xiong J, Rosen G, Gulley JL and Apolo AB. Avelumab in metastatic urothelial carcinoma after platinum failure (JAVELIN solid tumor): pooled results from two expansion cohorts of an open-label, phase 1 trial. Lancet Oncol 2018; 19: 51-64.

[169] Balar AV, Castellano D, O'Donnell PH, Grivas P, Vuky J, Powles T, Plimack ER, Hahn NM, de Wit R, Pang L, Savage MJ, Perini RF, Keefe SM, Bajorin D and Bellmunt J. First-line pembrolizumab in cisplatin-ineligible patients with locally advanced and unresectable or metastatic urothelial cancer (KEYNOTE-052): a multicentre, single-arm, phase 2 study. Lancet Oncol 2017; 18: 1483-1492.

[170] Bellmunt J, de Wit R, Vaughn DJ, Fradet Y, Lee JL, Fong L, Vogelzang NJ, Climent MA, Petrylak DP, Choueiri TK, Necchi A, Gerritsen W, Gurney H, Quinn DI, Culine S, Sternberg CN, Mai Y, Poehlein CH, Perini RF and Bajorin DF; KEYNOTE-045 Investigators. Pembrolizumab as second-line therapy for advanced urothelial carcinoma. N Engl J Med 2017; 376: 1015-1026.

[171] Loriot Y, Necchi A, Park SH, Garcia-Donas J, Huddart R, Burgess E, Fleming M, Rezazadeh A, Mellado B, Varlamov S, Joshi M, Duran I, Tagawa ST, Zakharia Y, Zhong B, Stuyckens K, Santiago-Walker A, De Porre P, O'Hagan A, Avadhani A and Siefker-Radtke AO; BLC2001 Study Group. Erdafitinib in locally advanced or metastatic urothelial carcinoma. N Engl J Med 2019; 381: 338-348.

[172] Petrylak DP, Perez RP, Zhang J, Smith DC, Ruether JD, Sridhar SS, Sangha RS, Lang JM, Heath EI, Merchan JR, Gartner EM, Chu R, Anand B, Doñate F, Jackson L, Adams J, Melhem-Bertrandt A and Rosenberg JE. A phase I study of enfortumab vedotin (ASG-22CE; ASG-22ME): updated analysis of patients with metastatic urothelial cancer. J Clin Oncol 2017; 35: 106-106.