PPM1G and its diagnostic, prognostic and therapeutic potential in HCC (Review)

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Abstract. Global statistics indicate that hepatocellular carcinoma (HCC) is the sixth most common cancer and the third leading cause of cancer-related death. Protein phosphatase Mg^{2+}/Mn^{2+} dependent 1G (PPM1G, also termed PP2Cγ) is one of the 17 members of the PPM family. The enzymatic activity of PPM1G is highly reliant on Mg^{2+} or Mn^{2+} and serves as a dephosphorylation regulator for numerous key proteins. PPM1G, functioning as a phosphatase, is involved in a number of significant biological processes such as the regulation of eukaryotic gene expression, DNA damage response, cell cycle and apoptosis, cell migration ability, cell survival and embryonic nervous system development. Additionally, PPM1G serves a role in regulating various signaling pathways. In recent years, further research has increasingly highlighted *PPM1G* as an oncogene in HCC. A high expression level of PPM1G is closely associated with the occurrence, progression and poor prognosis of HCC, offering notable diagnostic and therapeutic value for this patient population. In the present review, the regulatory role of PPM1G in diverse biological processes and signaling pathway activation in eukaryotes is evaluated. Furthermore, its potential application as a biomarker in the diagnosis and prognosis evaluation of HCC is assessed, and future prospects for HCC treatment strategies centered on PPM1G are discussed.

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Contents

1. Introduction

Primary liver cancer is the most frequently diagnosed malignant tumor of the liver, of which there are three main histological subtypes: Hepatocellular carcinoma (HCC), intrahepatic cholangiocarcinoma (ICC) and combined HCC-ICC (1), which account for ~80, 15 and 5% of cases, respectively (2). There are notable distinctions between the various histological subtypes in terms of epidemiology, clinicopathological morphology, genetic modifications and therapeutic responses (3). Globally, HCC is the sixth most prevalent disease and the third most common cause of cancer-related death, according to the GLOBOCAN 2020 statistics (4). The primary cause of HCC globally is liver cirrhosis from the hepatitis B virus (HBV) and hepatitis C virus (HCV) (5). A meta-analysis showed that HBV-related HCC is more common in Sub-Saharan Africa and South-East Asia, but HCV-related HCC is more common in North Africa and the West (6). Additionally, with the global increase in obesity and type 2 diabetes, non-alcoholic fatty liver disease and non-alcoholic steatohepatitis have become more prominent underlying causes of HCC (7). In recent decades, significant advancements have been achieved in the treatment of HCC through surgical interventions, chemotherapy, radiotherapy, targeted therapy, immunotherapy and other methods. However, the absence of early symptoms of HCC, coupled with the insufficient availability of diagnostic and prognostic biomarkers, continues to contribute to a poor 5-year survival rate among patients with advanced HCC (5).

Post-translational modification (PTM), which entails the addition of chemical groups to the amino acid side chains of proteins, serves a critical role in signal transduction, apoptosis, transcriptional regulation as well as cell-cell and cell-matrix interactions (8). Common PTMs include phosphorylation, acetylation, ubiquitination, glycosylation, amidation, hydroxylation and sumoylation (9-11), as well as the recently discovered histone lysine lactylation (12). Phosphorylation is the most common PTM, which controls almost all biological processes in eukaryotic cells (13,14). Protein phosphorylation is reversible and dynamic and is regulated by the opposing actions of protein kinases, which facilitate phosphorylation, and protein phosphatases, which catalyze phosphate removal (15). The three amino acids most commonly phosphorylated in proteins are serine (Ser), threonine (Thr) and tyrosine (14), among which Ser and Thr constituted >98% of all phosphorylated residues in human proteins (14). Metal-dependent protein phosphatase (PPM), also known as protein phosphatase 2C, is a highly conserved family of protein phosphatases (16,17). The common feature of PPM family members is a conserved catalytic core region, which contains metal chelating residues (18). Members of the PPM family are widely expressed in a number of different species (19). In *Homo sapiens*, there are at least 17 members of the PPM family, and it has been confirmed that certain members play a crucial role in cancer and disease-related physiological or pathological processes, such as transcriptional regulation, activation of signaling pathways, cell proliferation, invasion, migration, apoptosis and cell cycle regulation (19).

Over the past decade, a number of experimental and clinical studies have linked the subgroup, protein phosphatase Mg^{2+}/Mn^{2+} dependent 1G (PPM1G, also known as PP2Cγ), of the PPM family to the development and progression of HCC and support its use as a biomarker for HCC diagnosis and prognosis. The present review aims to provide an overview of the evidence that suggests PPM1G serves a functional role in the initiation and progression of human HCC. Furthermore, the prospective clinical utility of PPM1G as a biomarker for both diagnosing and prognosticating HCC, along with its potential as a novel therapeutic target for this disease is also evaluated.

2. Discovery and characterization of PPM1G

PPM1G, is a gene located in the short arm of human chromosome 2 (20), identified and characterized by Travis and Welsh (21) in 1997, which encodes a protein of 546 amino acids. Notably, although the predicted molecular weight of PPM1G is 59 kDa based on the amino acid composition, the observed molecular weight is 75 kDa (21). This discrepancy is due to the presence of a unique highly acidic (AD) domain comprising ~200 amino acids, a feature not found in other PPM phosphatases (21). Northern blotting analysis has shown that *PPM1G* is widely expressed in human tissues, and its transcription level peaks in human testis, skeletal muscle and heart tissues (21). In addition, as with the other known PPMs, the activity of PPM1G is highly dependent on Mg^{2+} or Mn^{2+} (17,21) (Fig. 1). It is speculated that there may be a link between PPM1G and cell proliferation (21). In 2021, Chen *et al* (22) were the first to utilize immunohistochemistry to validate that the expression of PPM1G was markedly upregulated in HCC tissues compared with adjacent tissues. The team also verified that PPM1G is associated with malignant biological processes such as the proliferation and invasion of HCC cell lines. Additionally, the expression level of PPM1G is significantly negatively correlated with the prognosis of patients with HCC following surgery (22).

3. Physiological functions of PPM1G

PPM1G regulates gene transcription elongation. Before genes are completely transcribed, RNA polymerase II (RNA-PII) interacts with the DRB sensitivity-inducing factor (DSIF) and negative elongation factor (NELF) in the proximal region of the promoter, resulting in a 'pause' in transcriptional elongation (23,24). CDK9, a key component of positive transcription elongation factor b (P-TEFb), phosphorylates Ser2 of the C-terminal domain of RNA-PII, as well as DSIF and NELF, leading to the release of RNA-PII from the proximal promoter and effectively terminating the transcriptional 'pause' state for efficient full-length mRNA synthesis (25,26). However, in eukaryotic cells, approximately half of P-TEFb molecules bind reversibly to HEXIM P-TEFb complex subunit 1 (HEXIM1) and are sequestered in the inactive 7SK small nuclear ribonucleoprotein (snRNP) complex (27).

McNamara *et al* (28,29) and other researchers have conducted extensive research on PPM1G-mediated transcriptional elongation regulation (30). In 2013, McNamara *et al* (28) utilized proteomics to identify an interaction between Tat (a key HIV transactivator of transcription) (31) and PPM1G. Subsequently, an enzymatic decomposition mechanism of 7SK snRNP mediated by PPM1G was proposed, which plays a crucial role in mRNA transcriptional elongation. The specific mechanism involves Tat recruiting PPM1G to the 7SK snRNP complex during HIV transcription. This recruitment leads to dephosphorylation of the T-loop of CDK9 by PPM1G, resulting in decomposition of the 7SK snRNP complex and the release of P-TEFb (28,32). Subsequently, Tat captures the dephosphorylated P-TEFb, stimulates autophosphorylation of the CDK9 T-loop and transfers the locally released kinase to the transactivating response region element (33) to facilitate the assembly of the transcription elongation complex (28). In 2015, Gudipaty *et al* (29) proposed that PPM1G could interact with the CR1 domain of HEXIM1 and bind to the 7SK RNA to create a 7SK-PPM1G snRNP complex. The formation of this complex therefore relieves the sequestering of P-TEFb caused by the formation of the 7SK snRNP complex by inhibiting the binding of P-TEFb to HEXIM1 (29) (Fig. 2A).

PPM1G regulates mRNA splicing. Precursor mRNA (pre-mRNA) splicing is a key step in the regulation of gene expression (34,35). Moreover, the formation of the A complex is generally considered to be the major determinant of splice site selection during alternative splicing as it typically enables the removal of introns (36). Previous studies have demonstrated that PPM1G can function as a pre-mRNA splicing factor, and its phosphatase activity and AD domain serve a crucial role in the formation of A complex (37,38). Furthermore, in an immunoprecipitation assay of an *in vitro* splicing reaction involving radiolabeled pre-mRNA, an anti-PPM1G monoclonal antibody efficiently precipitated spliced RNA precursors, intermediates

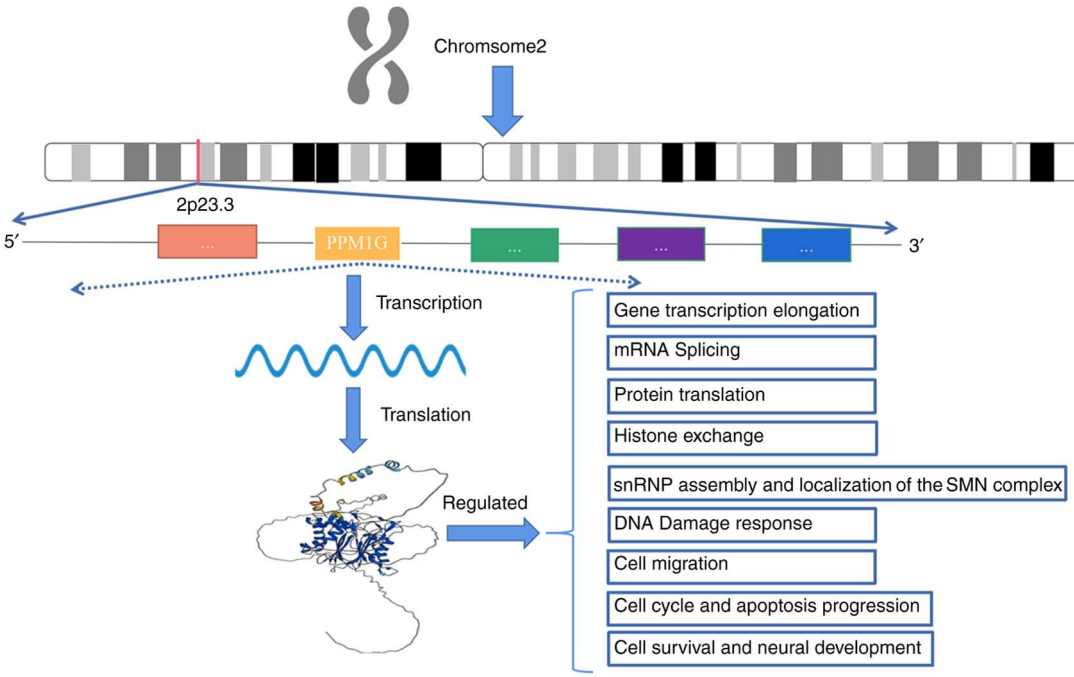

Figure 1. Schematic of PPM1G production. PPM1G, protein phosphatase Mg^{2+}/Mn^{2+} dependent 1G. SMN, survival of motor neurons; snRNP, small nuclear ribonucleoprotein.

and products (37). In summary, we consider that PPM1G serves a notable role in the whole splicing process (37). Moreover, PPM1G is regarded as a splicing factor that has an important role in regulating the alternative splicing of CD44 [Y-box binding protein 1 (YB-1) target] exons v4 and v5 by interacting with YB-1 (38).

PPM1G regulates protein translation. In eukaryotic cells, two protein synthesis mechanisms exist: The classic cap-dependent and the cap-independent protein translation mechanisms (39). Initiation of the cap-dependent protein translation mechanism involves the eukaryotic initiation factor 4F (eIF4F) complex recognizing and binding to the 5'-cap (7-methylguanosine nucleoside) domain of mRNA in a cap-dependent manner (40). Subsequently, the eIF4E subunit in the eIF4F complex recruits the 43S ribosomal subunit to the 5' end of the mRNA, thereby activating mRNA translation (40,41). A study by Liu *et al* (42) found that the protein synthesis inhibitor, 4E-BP1, is a direct substrate of PPM1G, and downregulation of PPM1G can significantly increase the phosphorylation level of four phosphorylation sites (Thr-37, Thr-46, Ser-65 and Thr-70) of 4E-BP1 (43). Additionally, a dual luciferase reporter gene assay demonstrated that PPM1G knockdown significantly enhances the cap-dependent translation process. A previous study has shown that dephosphorylated 4E-BP1 binds to the cap-binding subunit, eIF4E, in the eIF4F complex and isolates it from the cap initiation complex, thereby inhibiting the cap-dependent translation process (44). Therefore, it is postulated that PPM1G-mediated dephosphorylation of 4E-BP1 facilitates its binding to eIF4E within the cap-initiating complex, leading to the inhibition of protein translation and cell growth (Fig. 2B). Furthermore, Xu *et al* (45) first proposed in 2016 that activation of the PI-3K/AKT signaling pathway can lead to the phosphorylation of PPM1G, inhibiting its phosphatase activity and subsequently impeding its ability to dephosphorylate 4E-BP1, thereby activating the cap-dependent translation process.

PPM1G regulates histone exchange. Histone chaperones are a class of proteins with histone binding activity that can regulate the assembly and decomposition of nucleosomes, and which have been shown to serve a crucial role in promoting histone exchange (46-48). Kimura *et al* (49) proposed two potential histone exchange mechanisms involving PPM1G. First, PPM1G may interact with and modulate the phosphorylation status of Nap1/2, thereby regulating the chaperone activity of Nap1/2 in an ATP-independent manner and facilitating histone H2A-H2B exchange (50). Second, PPM1G can act as a histone chaperone by mediating the dephosphorylation of nucleosome-free H2A-H2B through its AD domain, subsequently incorporating H2A-H2B into chromatin (49).

PPM1G regulates localization of the survival of motor neurons (SMN) complex and snRNP assembly. The SMN complex is a stable multiprotein assembly composed of SMN, Gemin2-8 and Unrip (51,52). Unrip is primarily localized in the cytoplasm and is recruited into the SMN complex through its interaction with Gemin7 but it is not detected in the SMN complex of the Cajal bodies (CBs) and Gems in the nucleus (53,54). Petri *et al* (55) confirmed the specific binding between the SMN complex and PPM1G and found that knockdown of PPM1G expression significantly downregulated the expression levels of SMN and Gemin2 in cells, particularly in the nucleus. This indicates that PPM1G serves a prominent role in maintaining the stability of the SMN complex. It was further found that downregulation of nuclear PPM1G expression resulted in a significant increase in the phosphorylation

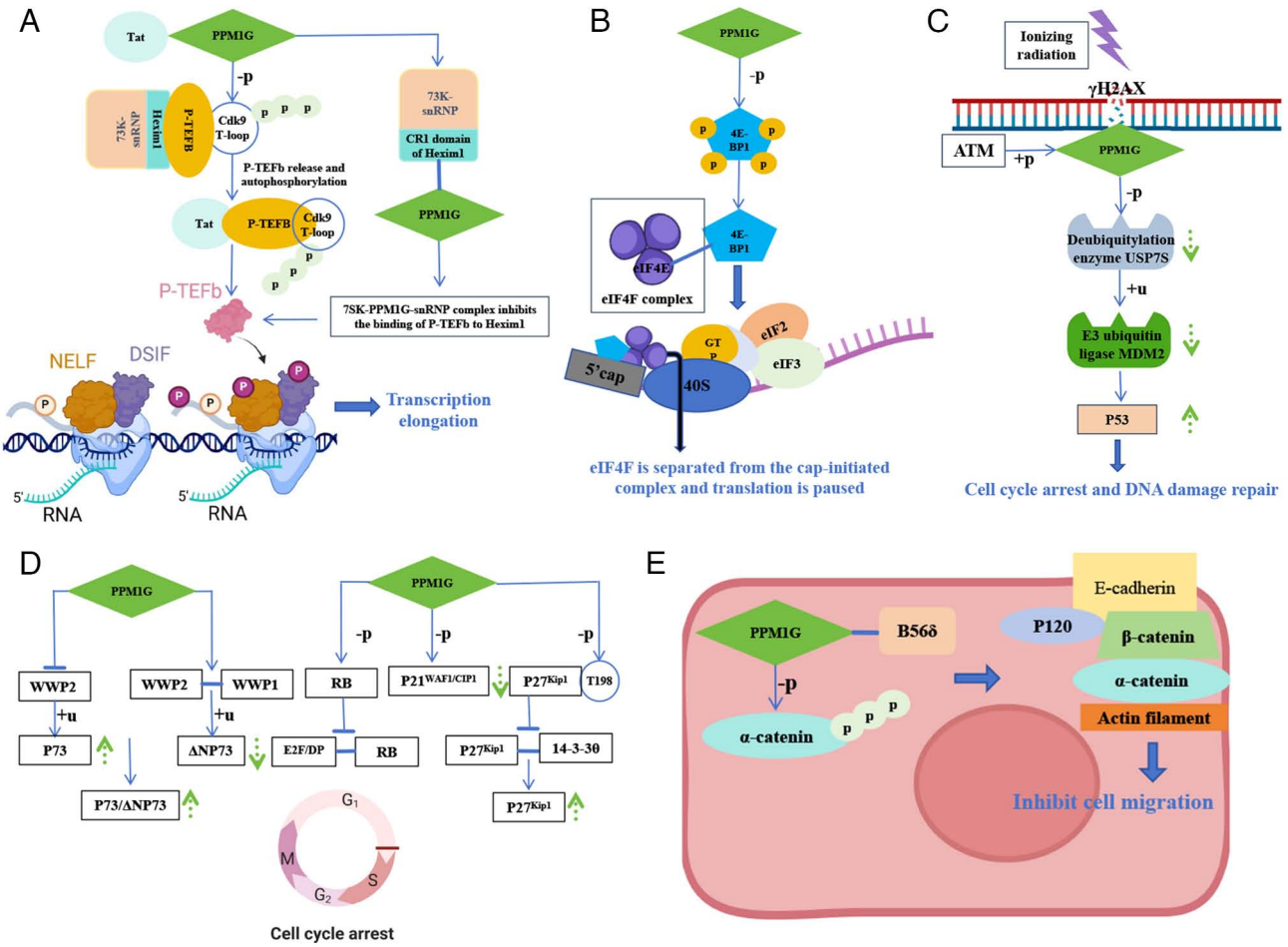

Figure 2. Regulatory role of PPM1G in a variety of biological processes. (A) PPM1G can dephosphorylate the T-loop of CDK9 or bind with HEXIM1 and 7SK RNA to create a 7SK-PPM1G snRNP complex. This process releases P-TEFb from the 7SK snRNP complex, thereby reactivating gene transcription elongation. (B) PPM1G can target and dephosphorylate 4E-BP1, and the dephosphorylated 4E-BP1 can then bind to the cap-binding subunit, eIF4E, in the eIF4F complex, which is subsequently isolated from the cap initiation complex, leading to a 'pause' in the protein translation process. (C) When ionizing radiation induces DNA damage, PPM1G is recruited to the DNA damage sites marked by phosphorylated H2AX. Subsequently, ATM phosphorylates and activates PPM1G. The activated PPM1G then targets and dephosphorylates the deubiquitinating enzyme, USP7S, leading to a series of cascading reactions, including a decrease in MDM2 levels and an increase in p53 levels. This results in cell cycle arrest, providing time for DNA repair and the restoration of genomic stability. (D) PPM1G can regulate the cell cycle and apoptosis processes in multiple ways: On the one hand, it specifically inhibits the E3 ligase activity of WWP2, promoting the assembly of the WWP2-WWP1 heterodimer complex, thereby maintaining the balance of cell cycle regulatory proteins, p73 and ΔNp73, within the cell. On the other hand, PPM1G can target and dephosphorylate the cell cycle regulatory proteins, RB, p21WAF1/CIP1 and p27Kip1, thereby inhibiting cell cycle progression. (E) The acidic domain of PPM1G can bind to B56δ, forming the PPM1G-B56δ holoenzyme complex, which in turn promotes the retention of PPM1G in the cytoplasm. The accumulation of this complex in the cytoplasm further enhances the specific dephosphorylation of α-catenin by PPM1G, facilitating the proper assembly of α-catenin and β-catenin in cells, and inhibiting cell migration. PPM1G, protein phosphatase Mg^{2+}/Mn^{2+} dependent 1G; CDK9, cyclin-dependent kinase 9; HEXIM1, HEXIM P-TEFb complex subunit 1; snRNP, small nuclear ribonucleoprotein; P-TEFb, positive transcription elongation factor b; 4E-BP1, 4E binding protein 1; eIF4E, eukaryotic initiation factor 4E; eIF4F, eukaryotic initiation factor 4F; ATM, ataxia-telangiectasia mutated; USP7S, USP7 deubiquitylation enzyme; MDM2, murine double minute 2; WWP1/2, WW domain containing E3 ubiquitin protein ligase 1/2.

levels of SMN and Gemin3 in the nucleus, as well as a reduction in the number of CBs in the nucleus (55). Previous studies have shown that cytoplasmic SMN is highly phosphorylated, while low phosphorylation contributes to the accumulation of SMN in the nucleus (55,56). In addition, changes in phosphorylation patterns can cause the SMN complex to translocate between the cytoplasm and the nucleus (56,57). Unrip, as a kinase, may be directly involved in the phosphorylation of cytoplasmic SMN complexes, and knockdown of Unrip can antagonize the change in subcellular localization of the SMN complex caused by PPM1G knockdown (55). In addition, the SMN complex, as a molecular chaperone, serves a notable role in the biogenesis and maturation of snRNP (58,59). The results of a band shift assay showed that the assembly of snRNPs was significantly reduced after knocking out PPM1G expression (55). This was considered to be closely related to the aforementioned downregulation of the stability of the SMN complex in the cytoplasm.

PPM1G regulates the DNA damage response (DDR). Ionizing radiation (IR) can cause DNA damage, particularly single-strand or double-strand breaks (DSBs), which are highly lethal (60-62). It has been established that phosphorylation of the C-terminal Ser residue (Ser139) in the histone H2A variant, H2AX, serves as a critical biomarker for DNA DSBs in mammalian cells (63). A study has shown that PPM1G is recruited to damaged DNA regions marked by phosphorylated H2AX during the DDR (64). Khoronenkova *et al* (65) found

that IR exposure leads to a decrease in the phosphorylation of the deubiquitylation enzyme, USP7S, accompanied by a decrease in murine double minute 2 (MDM2) and an increase in p53 levels. By contrast, PPM1G knockdown cells do not exhibit these effects (65). A previous study has shown that that USP7S colocalizes and interacts with MDM2 to reduce the destruction of the latter by proteasomes (65). The E3 ubiquitin ligase, MDM2 (a key negative regulator of p53), can bind to and ubiquitinate p53, leading to proteasomal degradation of p53 protein (66). When DNA damage occurs prior to cells entering the S phase, p53 may provide time for DNA repair and restoration of genomic stability by inducing cell cycle arrest in the G1 phase (67). In addition, genome-wide screening identified PPM1G as a potential target for phosphorylation by ataxia-telangiectasia mutated (ATM) (68). Furthermore, in ATM knockdown experiments, the dephosphorylation of USP7S was eliminated under IR conditions, indicating that the dephosphorylation of USP7S by PPM1G is dependent on ATM (65). The specific molecular mechanism involves DNA damage-induced ATM-mediated phosphorylation and subsequent activation of PPM1G. Once activated, PPM1G proceeds to dephosphorylate USP7S, thereby regulating its function (65,69). In addition, Khoronenkova *et al* (65) found that PPM1G knockdown significantly increased the expression of p21 protein in cells, and an increase in the proportion of cells arrested in the G1 phase of the cell cycle was detected. Notably, the expression level of p53 protein remained relatively stable during this process. An increased accumulation of unrepaired DNA strand breaks in the G1 phase was also observed, indicating that PPM1G-deficient cells can cause cell cycle arrest in a p53-independent manner, but cannot effectively repair endogenous DNA damage (65) (Fig. 2C).

PPM1G regulates cell cycle and apoptosis progression. Chaudhary and Maddika (70) confirmed the interaction between WW domain containing E3 ubiquitin protein ligase (WWP) 1 and WWP2 by Co-immunoprecipitation assay. Notably, it was also observed that the interaction between WWP1 and WWP2 was significantly attenuated following PPM1G knockdown (70). WWP2 and WWP1 are two crucial ubiquitin E3 ligases (71,72), that have pivotal roles in regulating the ubiquitination and subsequent proteasomal degradation of p73 and its isoform, ΔNp73, respectively (70). Furthermore, *in vitro* binding experiments demonstrated that the specific regulation of ΔNp73 ubiquitination levels by WWP1 is contingent upon the binding of WWP2 (70). As a sophisticated molecular switch, PPM1G exhibits dual functionality as it not only specifically inhibits the E3 ligase activity of WWP2 but also facilitates the assembly of the WWP2-WWP1 heterodimer complex (70). Therefore, through this intricate mechanism, PPM1G regulates the ubiquitin-proteasome pathway-mediated degradation of p73 and ΔNp73, thereby maintaining the balance of these proteins within cells (70). p73 is considered to have a highly similar structure and function to the p53 tumor suppressor protein, both of which serve a notable and decisive role in inducing apoptosis and cell cycle arrest (73,74).

Recent studies have revealed that the p53/p21/retinoblastoma (RB) protein signaling pathway plays a significant role in regulating the cell cycle and tumor inhibition (75,76). Suh *et al* (77) discovered that PPM1G may facilitate the degradation of $p21^{WAF1/CIP1}$ protein through its phosphatase activity and cause cell cycle arrest, which is manifested by an increase in the number of cells in the S phase. It has been confirmed that this regulatory mechanism is independent of p53 (77). In addition, further experiments demonstrated that overexpression of PPM1G resulted in a decrease in the phosphorylation level of the RB protein in 293 cells, while its overall expression remained relatively constant (77). Previous research has demonstrated that the RB protein, functioning as a tumor suppressor, inhibits S phase entry and cellular proliferation by interacting with members of the E2F/DP transcription factor family (78,79). The phosphorylation of RB can result in its functional inactivation (79). Hence, it is suggested that PPM1G also has a crucial role in cell cycle modulation by dephosphorylating RB (77).

$p27^{Kip1}$ is a cyclin-dependent kinase inhibitor that has been demonstrated to serve a crucial inhibitory role in the transition from the G1 to the S phase of the cell cycle (80-82). When $p27^{Kip1}$ is phosphorylated, the cell cycle negative regulator, 14-3-3θ, can bind to it, causing it to translocate to the cytoplasm and be degraded (83). Sun *et al* (84) discovered that PPM1G is capable of dephosphorylating the Thr198 residue of $p27^{Kip1}$, thereby inhibiting its interaction with 14-3-3θ and reducing the degradation of $p27^{Kip1}$ (Fig. 2D).

PPM1G regulates cell migration. Changes in intercellular and cell-extracellular matrix adhesion are considered key steps leading to alterations in the nature of tumor metastasis and invasion (85). Both the N and C termini of α-catenin can bind to β-catenin and actin filaments and play a notable role in intercellular connections (86-88). α-catenin downregulation can result in the loss of cell-cell adhesion ability and promotes malignant biological processes such as tumor cell invasion and migration (86). In a 2019 study, Kumar *et al* (89) found that the interaction between α-catenin and β-catenin occurs in the cytoplasm and nucleus, while the specific interaction between α-catenin and PPM1G only occurs in the cytoplasm. It was also found that B56δ can bind to the AD domain of PPM1G, forming a PPM1G-B56δ holoenzyme complex, which results in the retention of PPM1G within the cytoplasm (89). The accumulation of this complex in the cytoplasm in turn significantly enhances the specific dephosphorylation of cytoplasmic α-catenin by PPM1G, promotes the proper assembly of α-catenin and β-catenin in cells and inhibits cell migration (89) (Fig. 2E).

PPM1G regulates cell survival and neural development. Foster *et al* (90) found that PPM1G is highly expressed in the embryonic neural structure of vertebrates by X-gal staining and demonstrated that PPM1G is essential for the survival of embryos. Further studies have found that normal expression of PPM1G not only regulates the resistance of cells to genotoxic stress and oxidative stress during development but can also inhibit the activation of the p38 MAPK pro-apoptotic stress signaling pathway (described further below) (90,91). Significant defects in central nervous system development in PPM1G-knockdown zebrafish have also been observed (90). Thus, PPM1G is regarded as a key regulator in cell survival and neural development (90).

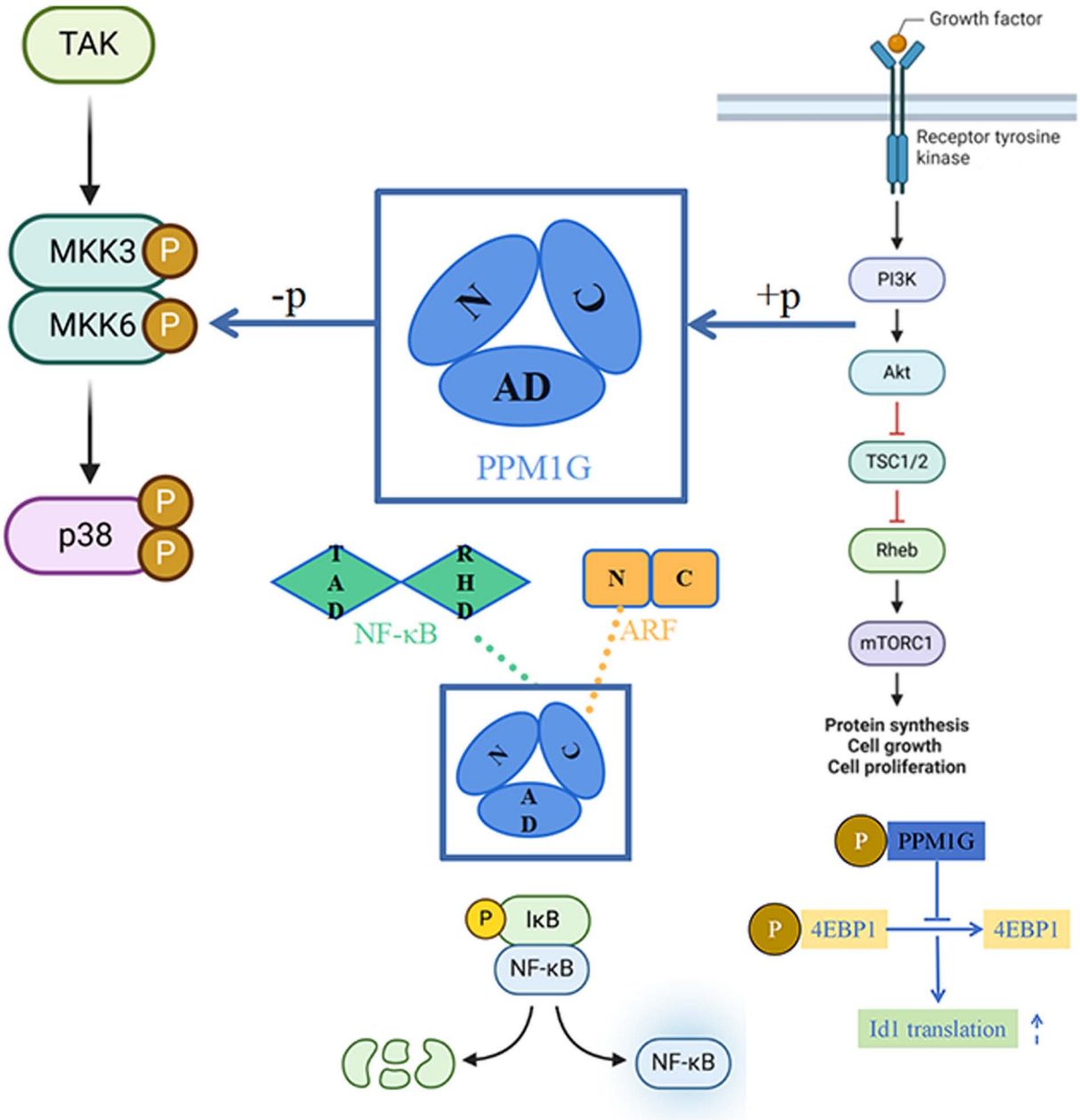

Figure 3. PPM1G-associated signaling pathways. PPM1G, protein phosphatase Mg^{2+}/Mn^{2+} dependent 1G; 4E-BP1, 4E binding protein 1; MKK, MAPK kinase; ARF, ADP ribosylation factor; TSC1/2, tuberous sclerosis proteins 1/2; 4E-BP1, 4E binding protein 1; p, phosphorylation.

4. PPM1G-associated signaling pathways

p38 MAPK signaling pathway. In the p38 MAPK signaling pathway, p38 MAPK is activated by upstream MAPK kinases (MKKs) such as MKK3 and MKK6 (92,93). The activation of p38 MAPK inhibits cell apoptosis, cell growth and differentiation (94). As a phosphatase, PPM1G can promote the dephosphorylation of MKK6, thereby inhibiting the activity of p38 (91). Through these mechanisms, PPM1G can facilitate the proliferation, invasion, migration and other malignant biological processes of lung adenocarcinoma (91) (Fig. 3).

Nuclear factor (NF)-κB signaling pathway. One of the most frequently implicated human cancer genes in different patient age groups and tumor types is INK4a/ADP ribosylation factor (ARF) (95). After stimulation with pro-inflammatory cytokines (such as tumor necrosis factor-α), the master transcription regulator, NF-κB, utilizes PPM1G as a coactivator to induce inflammation and normal cell survival programs (96). Additionally, PPM1G can assemble a ternary protein complex by bridging the N-terminus of ARF and the Rel homology domain of NF-κB, thereby mediating the interaction between ARF and NF-κB (96). The binding of ARF with PPM1G not only directly reduces NF-κB activity, but also acts as a negative regulatory factor for PPM1G-dependent NF-κB transcription to regulate processes such as inflammation and cell apoptosis, thereby serving a role in the occurrence and development of various tumor-related diseases (96). Finally, a study found that NF-κB can play a role similar to Tat [recruits PPM1G to the promoter of inflammatory response genes] and utilize P-TEFb activation to enhance the transcriptional extension and activation of inflammatory response genes (28) (Fig. 3).

PI-3K/AKT/mTOR signaling pathway. It has been well-established that dephosphorylated 4E-BP1 inhibits cap-dependent translation by binding to the translation initiation factor, eIF4E (44). Xu *et al* (45) first proposed in 2016 that activation of the PI-3K/AKT signaling pathway can regulate the expression of the Inhibitors of DNA binding and cell differentiation 1 (Id1) protein at the post-transcriptional level (97). First, activation of the PI-3K/AKT signaling pathway can lead to the phosphorylation of 4E-BP1, which subsequently impedes its interaction with eIF4E, thereby activating the cap-dependent translation process of Id1 protein (45). Second, PPM1G is phosphorylated in a PI-3K/AKT-dependent manner, resulting in decreased phosphatase activity, increased phosphorylation of 4E-BP1 and activation of Id1 translation, resulting in increased Id1 expression (45) (Fig. 3).

5. PPM1G and its diagnostic and prognostic potential in HCC

Previous studies have indicated that the expression level of PPM1G in liver cancer samples is significantly higher than that in normal liver tissue (22,98,99). The potential of PPM1G in HCC diagnosis and prognosis is gradually being recognized. Hu *et al* (99) conducted immunohistochemical staining of 96 HCC tumor and adjacent tissues samples obtained from the Cancer Center of Sun Yat-sen University (Guangzhou, China). The expression levels of PPM1G in these tissues were quantified and these findings were correlated with the patient clinicopathological parameters and prognostic information. The analysis revealed that high PPM1G expression was significantly associated with elevated serum α-fetoprotein levels (P=0.015), advanced clinical stage (P=0.035), poor differentiation (P=0.027), increased cancer embolus (P=0.022) and tumor recurrence (P=0.015) in patients with HCC (99). Furthermore, Kaplan-Meier survival analysis demonstrated that patients with higher PPM1G expression had a poorer overall survival (OS; P<0.0001) and disease-free survival (P=0.004) (99). These findings align with the conclusions reported by Xiong *et al* (98) in 2022. Additionally, Xiong *et al* (98) demonstrated that the PPM1G mRNA expression level in patients with HCC was positively correlated with TNM stage, tumor grade and metastasis. Notably, Cox multivariate regression analysis revealed that the PPM1G expression level (P=0.03) was an independent risk factor influencing the prognosis of patients with HCC (98,100).

Liu *et al* (100) utilized The Cancer Genome Atlas public database to categorize HCC patients undergoing sorafenib treatment into responders and non-responders. By comparing the differentially expressed genes between these two groups, a total of 393 sorafenib-responsive target genes were identified in HCC tissues, with 352 of these target genes being upregulated. Subsequently, through Kaplan-Meier survival and receiver operating characteristic curve analyses, five sorafenib-responsive target genes [*SLC41A3*, *SEC61A1*, *LRP4*, *PPM1G* and *HSP90AA1*] were identified as highly expressed in HCC tissues and significantly correlated with both prognosis and diagnosis. Additionally, univariate and multivariate Cox regression analyses indicated that these five candidate genes were independent factors contributing to the poor prognosis of patients with HCC (100). Based on these findings, the research team developed a risk model based on the sorafenib-responsive target genes. In addition, Liu *et al* (100) also demonstrated that the risk model based on sorafenib-response target genes [*SLC41A3*, *SEC61A1*, *LRP4*, *PPM1G*, *HSP90AA1*] was highly correlated with the infiltration level of immune cells in HCC.

In 2022, Li *et al* (101) utilized bioinformatics analysis to identify five ferroptosis-related genes [*PPM1G*, *MED8*, *PIGU*, *RAN* and *SNRPB*] associated with the onset and progression of HCC as well as patient prognosis. These genes were subsequently utilized to develop a multi-gene risk prediction model for HCC, where a higher risk score was linked to a poorer OS in patients. Additionally, Li *et al* (101) confirmed a significant correlation between the risk score of the model and the TNM stage (Table I).

As is well-known, the occurrence of HCC is relatively hidden, with a notable absence of specific clinical symptoms in its early stages. It is therefore of great importance to identify a diagnostic marker with high sensitivity and specificity for the diagnosis of this disease. However, traditional single-marker approaches often fall short in meeting the demands of clinical applications. The utilization of a comprehensive panel of multiple markers significantly enhances the detection rate of early-stage cancer. In recent years, researchers have utilized various research methods to discover that PPM1G is not only a target of the therapy drug, sorafenib (100), but also exhibits a strong correlation with ferroptosis (101). Building on these findings, several HCC multi-gene diagnosis and prognosis prediction models have been developed (100-102). Notably, these models demonstrate superior diagnostic and prognostic evaluation capabilities compared with traditional biomarkers and pathological staging systems, such as TNM (102-104) (Table I). Therefore, it is of paramount importance to further evaluate the diagnostic and prognostic efficacy of these models in clinical practice for patients with HCC, and to facilitate the clinical application transformation of these models.

6. Molecular mechanism of PPM1G in promoting HCC development and its prospect in HCC treatment

Chen *et al* (22) also found a significant negative correlation between the PPM1G expression level and the prognosis of patients with HCC. Knockdown of PPM1G resulted in a significant inhibition of the proliferation and invasion of HepG2 and Hep3B cells. This study also investigated the role of PPM1G in a mouse xenograft tumor model. The results demonstrated that mice with high PPM1G expression levels exhibited larger subcutaneous tumor volumes and weights compared with mice with low expression levels (22). The team also identified a distinct interaction between PPM1G and Ser/arginine-rich splicing factor 3 (SRSF3) through proteomics analysis and immunoprecipitation experiments. SRSF3 is a crucial multifunctional splicing factor that has a significant role in RNA biogenesis and processing, including in gene transcription and RNA splicing regulation (105). As a potential inhibitor of HCC (106), the role of SRSF3 in enhancing the sensitivity of HCC radiotherapy has been validated (107). The potential mechanism involves the activation of multiple transcription factors and co-activators during the onset and progression of HCC, leading to the upregulation of PPM1G expression (22). PPM1G interacts with SRSF3,

Table I. Hepatocellular carcinoma risk prediction model and its prognostic value.

Parameter	Yang *et al*, 2021 (102)	Li *et al*, 2022 (101)
Gene composition	*PPM1G, SRXN1, LDHA, TFDP1 and EIF2S1*	*PPM1G, MED8, PIGU, RAN and SNRPB*
AUC values for predicting OS in TCGA cohort		
0.5 years	Not measured	0.764
1 years	0.838	0.768
2 years	Not measured	0.712
3 years	0.741	0.722
5 years	0.692	0.702
AUC values for predicting OS in ICGC cohort		
0.5 years	Not measured	0.725
1 years	0.746	0.746
2 years	Not measured	0.801
3 years	0.778	0.823
5 years	0.778	0.785

AUC, area under the curve; EIF2S1, Eukaryotic translation initiation factor 2 subunit 1; ICGC, International Cancer Genome Consortium; LDHA, lactate dehydrogenase A; MED8, mediator complex subunit 8; OS, overall survival; PIGU, phosphatidylinositol glycan anchor biosynthesis class U; PPM1G, protein phosphatase Mg^{2+}/Mn^{2+} dependent 1G; SNRPB, mall nuclear ribonucleoprotein polypeptides B and B1; SRXN1, sulfiredoxin 1; TCGA, The Cancer Genome Atlas; TFDP1, transcription factor Dp-1.

promoting its dephosphorylation. The dephosphorylation of SRSF3 results in its dissociation from the pre-mRNA of genes related to cell proliferation, migration, cell cycle and transcriptional regulation, such as caspase 8 and protein arginine methyltransferase 1. This results in SRSF3 losing its ability to regulate the alternative splicing of these pre-mRNAs, thereby promoting malignant biological phenotypes such as tumor cell proliferation and invasion (22) (Fig. 4 and Table II).

As described in the previous section, Hu *et al* (99) confirmed that PPM1G expression levels were significantly higher in HCC tissues and cell lines than in adjacent tissues and normal liver cells, and that high PPM1G expression often indicates that patients with HCC will have poor clinical outcomes. *TP53* is a crucial tumor suppressor gene and the most frequently mutated gene in human cancer (108). The majority of *TP53* mutations in cancer are missense mutations that result in the production of a full-length mutant p53 protein (109). This mutated p53 protein not only loses its original tumor suppressor function but also gains oncogenic activity independent of wild-type p53, a phenomenon known as gain-of-function (GOF) (109). A study has revealed a clear interaction between PPM1G and the GOF mutant p53. Specifically, in cells overexpressing PPM1G, the expression level of GOF mutant p53 protein was significantly elevated (99). This may be related to the aforementioned regulation of the PPM1G/USP7S/MDM2 axis (65). In addition, the study demonstrated that PPM1G significantly enhanced the proliferation of HCC cells via both *in vitro* and *in vivo* experiments (99). However, a limitation of this study is that the specific mechanism of action of the PPM1G/GOF mutant p53 axis in the occurrence and development of HCC was not further verified (99). However, a recent study by Zhao *et al* (110) reported that GOF mutant p53 can promote tumor cell invasion and metastasis through the minichromosome maintenance complex component 5/chromosomal instability-cytoplasmic DNA/cyclic GMP-AMP synthase/stimulator of interferon genes (STING)-induced non-canonical NF-κB signaling pathway. Based on these findings, the team hypothesized that PPM1G may enhance the proliferation of HCC cells by modulating the expression and accumulation of GOF mutant p53 protein in HCC (99) (Fig. 4 and Table II).

It is well known that fibrosis is a key risk factor for liver disease and HCC, and hepatic stellate cells (HSCs) and macrophages are the main functional cell types in the liver fibrosis process (111). The Notch signaling pathway plays a notable role in HSC activation and macrophage M1 polarization (111). It has been reported that costunolide (COS), a natural sesquiterpene lactone isolated from *Saussurea lappa* (Compositae), can significantly reduce CCl_4 and bile duct ligation-induced liver injury and liver fibrosis in rats (112). The specific mechanism involves COS blocking the inhibitory effect of PPM1G on WWP2 by interfering with the interaction between PPM1G and WWP2, making Notch3 unstable (70). This inhibition results in the suppression of the activation of the Notch3/hes family bHLH transcription factor 1 (HES1) signaling pathway, inhibiting the liver fibrosis process (112) (Fig. 4 and Table II).

According to the Barcelona Clinic Liver Cancer classification, hepatectomy and transplantation are two of the most effective treatments for patients with HCC (113). Hepatic ischemia-reperfusion injury (IRI) is characterized by the activation of inflammatory networks and oxidative stress in tissues following reperfusion after a period of insufficient

Table II. Expression and roles of PPM1G in liver fibrosis and liver cancer.

First author, year	Pathology type	Expression	Related genes and signaling pathways	Functional roles	(Refs.)
Ge *et al*, 2020	Liver fibrosis	Upregulated	WWP2 and Notch3/HES1 signaling pathway	Promoting the progress of liver fibrosis.	(112)
Chen *et al*, 2021	Liver cancer	Upregulated	SRSF3	Promoting cell proliferation, invasion.	(22)
Hu *et al*, 2024	Liver cancer	Upregulated	p53	Promoting cell proliferation or liver transplantation.	(99)
Peng *et al*, 2024	Liver cancer	Upregulated	STING signaling pathway	Inhibition of IRI after hepatectomy.	(115)

HES1, hes family bHLH transcription factor 1; IRI, ischemia-reperfusion injury; PPM1G, protein phosphatase Mg^{2+}/Mn^{2+} dependent 1G; SRSF3, Ser/arginine-rich splicing factor 3; STING, stimulator of interferon genes; WWP2, WW domain containing E3 ubiquitin protein ligase 2.

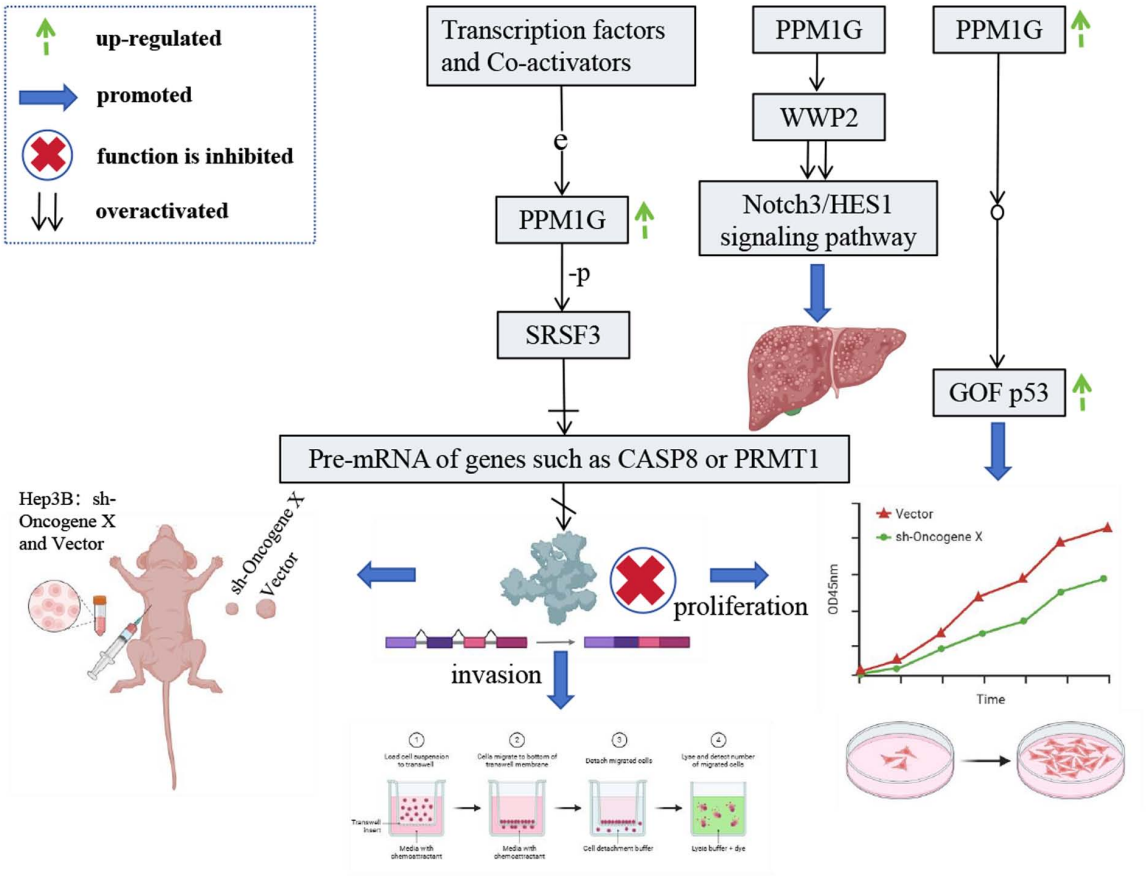

Figure 4. PPM1G participates in the occurrence and development of hepatocellular carcinoma and liver fibrosis via different pathways. i) In the occurrence and development of HCC, multiple transcription factors and co-activators are activated, promoting the expression of PPM1G. PPM1G can target and dephosphorylate SRSF3, leading to the dissociation of SRSF3 from the pre-mRNA of CASP8 and PRMT1. Consequently, SRSF3 loses its ability to regulate the alternative splicing of these pre-mRNAs, thereby promoting the proliferation and invasion of HCC cells. ii) PPM1G can promote the proliferation of HCC cells by enhancing the expression of GOF mutant p53. iii) PPM1G can target and inhibit WWP2, which leads to the overactivation of the Notch3/HES1 signaling pathway, promoting the development of liver fibrosis. HCC, hepatocellular carcinoma; PPM1G, protein phosphatase Mg^{2+}/Mn^{2+} dependent 1G; SRSF3, Ser/arginine-rich splicing factor 3; PRMT1, protein arginine methyltransferase 1; GOF mutant p53, gain-of-function mutant p53; WWP2, WW domain containing E3 ubiquitin protein ligase 2; HES1, hes family bHLH transcription factor 1; CASP8, caspase 8; sh, short hairpin.

oxygen supply, leading to cellular death (114). This condition is recognized as one of the most common and severe complications that adversely affects patient outcomes post-hepatectomy and transplantation (114). Upregulation of PPM1G inhibits activation of the STING signaling pathway and the release of inflammatory cytokines and can limit the polarization of

macrophages to the M1 phenotype (115). The dephosphorylation of STING by PPM1G has been reported as early as 2020 (116). It has been reported that the envelope protein, ORF33, of Kaposi's sarcoma-associated herpesvirus can promote the dephosphorylation of STING and mitochondrial antiviral signaling (MAVS) protein by recruiting PPM1G. This action limits the natural immune signals mediated by STING and MAVS, inhibits the antiviral activity of the host and thereby promotes virus lysis and replication (116). It has also been observed that knockdown of PPM1G significantly aggravates the liver IRI response in established 70% warm liver I/R mouse models (a classic *in vitro* liver IRI model) (115). Therefore, PPM1G, as a protective factor, has a crucial role in limiting I/R-mediated liver injury (115) (Table II).

The mechanism of action of tumor-specific antigen mRNA vaccines involves activating the immune system to target tumor cells using synthetic mRNA molecules, eliciting a broad spectrum of immune responses and reducing tumor evasion (117,118). Due to its high specificity, efficacy and minimal side effects, this approach holds promise in the realm of cancer therapy (119). A positive correlation between PPM1G expression and the presence of macrophages, dendritic cells and infiltrating B cells has been observed (120). Furthermore, the PPM1G tumor-specific antigen can be recognized by antigen-presenting cells, presented to B cells and stimulate lymphocyte infiltration in the tumor microenvironment, thereby triggering an immune response (120). Therefore, PPM1G is a promising candidate antigen for the development of mRNA vaccines that target HCC (120). These findings highlight the far-reaching significance of the abnormally upregulated oncogene, PPM1G, in HCC tumors for the diagnosis, prognosis evaluation and treatment of HCC. Regretfully, to the best of our knowledge, no clinical trials that address PPM1G as a treatment target or biomarker for HCC have been conducted.

7. Conclusions

HCC is a highly heterogeneous tumor caused by multiple gene mutations; therefore, the efficacy of treatment based on a single therapeutic target is often limited. PPM1G appears to demonstrate great potential in the treatment of HCC. On the one hand, as a tumor-promoting factor, PPM1G not only enhances HCC cell proliferation and invasion by modulating SRSF3 phosphorylation and increasing GOF mutant p53 protein expression but can also promote the occurrence of liver fibrosis by overactivating the Notch3/HES1 signaling pathway. On the other hand, analysis has shown that PPM1G was also significantly correlated with immune cell infiltration in HCC tissues. Therefore, targeting PPM1G can also attack tumors through tumor immunotherapy. Notably, studies have confirmed that PPM1G can regulate a variety of biological processes and signaling pathways related to the occurrence and development of human diseases. We consider that the specific mechanism of PPM1G in the occurrence and development of HCC has not been fully analyzed. Therefore, the safety and efficacy of targeted therapy based on PPM1G requires further basic and clinical experiments to accumulate more evidence. In anticipation of the future, our goal is to develop practical and effective targeted therapeutic strategies for HCC based on the specific molecular mechanisms involved. Concurrently, another research direction is to advance the clinical implementation of established diagnostic and prognostic evaluation models, to enable early detection and intervention at the secondary prevention stage of neoplastic diseases. Through this, we aim to bring hope to more patients with HCC.

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Authors' contributions

XZ drafted and revised the manuscript. HW, YY, JZ and JY collected the relevant papers and helped to revise the manuscript. XZ and HW designed the tables and charts. JH and LZ reviewed the article. Data authentication is not applicable. All authors read and approved the final version of the manuscript.

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Competing interests

The authors declared that they have no competing interests.

References

1. Zhang X, Wang Z, Tang W, Wang X, Liu R, Bao H, Chen X, Wei Y, Wu S, Bao H, *et al*: Ultrasensitive and affordable assay for early detection of primary liver cancer using plasma cell-free DNA fragmentomics. Hepatology 76: 317-329, 2022.
2. Rumgay H, Ferlay J, de Martel C, Georges D, Ibrahim AS, Zheng R, Wei W, Lemmens VEPP and Soerjomataram I: Global, regional and national burden of primary liver cancer by subtype. Eur J Cancer 161: 108-118, 2022.
3. Feng M, Pan Y, Kong R and Shu S: Therapy of primary liver cancer. Innovation (Camb) 1: 100032, 2020.
4. Sung H, Ferlay J, Siegel RL, Laversanne M, Soerjomataram I, Jemal A and Bray F: Global cancer statistics 2020: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. CA Cancer J Clin 71: 209-249, 2021.
5. Rumgay H, Arnold M, Ferlay J, Lesi O, Cabasag CJ, Vignat J, Laversanne M, McGlynn KA and Soerjomataram I: Global burden of primary liver cancer in 2020 and predictions to 2040. J Hepatol 77: 1598-1606, 2022.
6. Stroffolini T and Stroffolini G: A historical overview on the role of hepatitis B and C viruses as aetiological factors for hepatocellular carcinoma. Cancers (Basel) 15: 2388, 2023.

7. Foerster F, Gairing SJ, Müller L and Galle PR: NAFLD-driven HCC: Safety and efficacy of current and emerging treatment options. J Hepatol 76: 446-457, 2022.
8. Huang KY, Su MG, Kao HJ, Hsieh YC, Jhong JH, Cheng KH, Huang HD and Lee TY: dbPTM 2016: 10-Year anniversary of a resource for post-translational modification of proteins. Nucleic Acids Res 44 (D1): D435-D446, 2016.
9. Ebert T, Tran N, Schurgers L, Stenvinkel P and Shiels PG: Ageing-oxidative stress, PTMs and disease. Mol Aspects Med 86: 101099, 2022.
10. Zafar S, Fatima SI, Schmitz M and Zerr I: Current technologies unraveling the significance of post-translational modifications (PTMs) as crucial players in neurodegeneration. Biomolecules 14: 118, 2024.
11. Wang Y, Liu Z, Bian X, Zhao C, Zhang X, Liu X and Wang N: Function and regulation of ubiquitin-like SUMO system in heart. Front Cell Dev Biol 11: 1294717, 2023.
12. Chen L, Huang L, Gu Y, Cang W, Sun P and Xiang Y: lactate-lactylation hands between metabolic reprogramming and immunosuppression. Int J Mol Sci 23: 11943, 2022.
13. Safabakhsh S, Panwar P, Barichello S, Sangha SS, Hanson PJ, Petegem FV and Laksman Z: The role of phosphorylation in atrial fibrillation: A focus on mass spectrometry approaches. Cardiovasc Res 118: 1205-1217, 2022.
14. Song W, Hu L, Ma Z, Yang L and Li J: Importance of tyrosine phosphorylation in hormone-regulated plant growth and development. Int J Mol Sci 23: 6603, 2022.
15. Stanford SM and Bottini N: Targeting protein phosphatases in cancer immunotherapy and autoimmune disorders. Nat Rev Drug Discov 22: 273-294, 2023.
16. Gao C, Cao N and Wang Y: Metal dependent protein phosphatase PPM family in cardiac health and diseases. Cell Signal 85: 110061, 2021.
17. Kamada R, Kudoh F, Ito S, Tani I, Janairo JIB, Omichinski JG and Sakaguchi K: Metal-dependent Ser/Thr protein phosphatase PPM family: Evolution, structures, diseases and inhibitors. Pharmacol Ther 215: 107622, 2020.
18. Das AK, Helps NR, Cohen PT and Barford D: Crystal structure of the protein serine/threonine phosphatase 2C at 2.0 A resolution. EMBO J 15: 6798-6809, 1996.
19. Pan C, Tang JY, Xu YF, Xiao P, Liu HD, Wang HA, Wang WB, Meng FG, Yu X and Su JP: The catalytic role of the M2 metal ion in PP2Cα. Sci Rep 5: 8560, 2015.
20. The GeneCards human gene database: Weizmann Institute of Science; [updated 30-5-2024]. Available from: www.genecards.org/Search/Keyword?queryString=PPM1G.
21. Travis SM and Welsh MJ: PP2C gamma: A human protein phosphatase with a unique acidic domain. FEBS Lett 412: 415-419, 1997.
22. Chen D, Zhao Z, Chen L, Li Q, Zou J and Liu S: PPM1G promotes the progression of hepatocellular carcinoma via phosphorylation regulation of alternative splicing protein SRSF3. Cell Death Dis 12: 722, 2021.
23. Mandal R, Becker S and Strebhardt K: Targeting CDK9 for anti-cancer therapeutics. Cancers (Basel) 13: 3181, 2021.
24. Whelan M and Pelchat M: Role of RNA polymerase II promoter-proximal pausing in viral transcription. Viruses 14: 2029, 2022.
25. Tellier M, Zaborowska J, Neve J, Nojima T, Hester S, Fournier M, Furger A and Murphy S: CDK9 and PP2A regulate RNA polymerase II transcription termination and coupled RNA maturation. EMBO Rep 23: e54520, 2022.
26. Fujinaga K, Huang F and Peterlin BM: P-TEFb: The master regulator of transcription elongation. Mol Cell 83: 393-403, 2023.
27. Yamayoshi A, Fukumoto H, Hayashi R, Kishimoto K, Kobori A, Koyanagi Y, Komano JA and Murakami A: Development of 7SK snRNA mimics that inhibit HIV transcription. ChemMedChem 16: 3181-3184, 2021.
28. McNamara RP, McCann JL, Gudipaty SA and D'Orso I: Transcription factors mediate the enzymatic disassembly of promoter-bound 7SK snRNP to locally recruit P-TEFb for transcription elongation. Cell Rep 5: 1256-1268, 2013.
29. Gudipaty SA, McNamara RP, Morton EL and D'Orso I: PPM1G binds 7SK RNA and Hexim1 to block P-TEFb assembly into the 7SK snRNP and sustain transcription elongation. Mol Cell Biol 35: 3810-3828, 2015.
30. Gudipaty SA and D'Orso I: Functional interplay between PPM1G and the transcription elongation machinery. RNA Dis 3: e1215, 2016.
31. Bagashev A and Sawaya BE: Roles and functions of HIV-1 Tat protein in the CNS: an overview. Virol J 10: 358, 2013.
32. Mbonye U, Wang B, Gokulrangan G, Shi W, Yang S and Karn J: Cyclin-dependent kinase 7 (CDK7)-mediated phosphorylation of the CDK9 activation loop promotes P-TEFb assembly with Tat and proviral HIV reactivation. J Biol Chem 293: 10009-10025, 2018.
33. Schulze-Gahmen U and Hurley JH: Structural mechanism for HIV-1 TAR loop recognition by Tat and the super elongation complex. Proc Natl Acad Sci USA 115: 12973-12978, 2018.
34. Hluchý M, Gajdušková P, Ruiz de Los Mozos I, Rájecký M, Kluge M, Berger BT, Slabá Z, Potěšil D, Weiß E, Ule J, *et al*: CDK11 regulates pre-mRNA splicing by phosphorylation of SF3B1. Nature 609: 829-834, 2022.
35. Borišek J, Casalino L, Saltalamacchia A, Mays SG, Malcovati L and Magistrato A: Atomic-level mechanism of pre-mRNA splicing in health and disease. Acc Chem Res 54: 144-154, 2021.
36. Ule J and Blencowe BJ: Alternative splicing regulatory networks: Functions, mechanisms, and evolution. Mol Cell 76: 329-345, 2019.
37. Murray MV, Kobayashi R and Krainer AR: The type 2C Ser/Thr phosphatase PP2Cgamma is a pre-mRNA splicing factor. Genes Dev 13: 87-97, 1999.
38. Allemand E, Hastings ML, Murray MV, Myers MP and Krainer AR: Alternative splicing regulation by interaction of phosphatase PP2Cgamma with nucleic acid-binding protein YB-1. Nat Struct Mol Biol 14: 630-638, 2007.
39. Wu S and Wagner G: Computational inference of eIF4F complex function and structure in human cancers. Proc Natl Acad Sci USA 121: e2313589121, 2024.
40. Jia X, He X, Huang C, Li J, Dong Z and Liu K: Protein translation: Biological processes and therapeutic strategies for human diseases. Signal Transduct Target Ther 9: 44, 2024.
41. Thompson L, Depledge DP, Burgess HM and Mohr I: An eIF3d-dependent switch regulates HCMV replication by remodeling the infected cell translation landscape to mimic chronic ER stress. Cell Rep 39: 110767, 2022.
42. Liu J, Stevens PD, Eshleman NE and Gao T: Protein phosphatase PPM1G regulates protein translation and cell growth by dephosphorylating 4E binding protein 1 (4E-BP1). J Biol Chem 288: 23225-23233, 2013.
43. Gingras AC, Raught B, Gygi SP, Niedzwiecka A, Miron M, Burley SK, Polakiewicz RD, Wyslouch-Cieszynska A, Aebersold R and Sonenberg N: Hierarchical phosphorylation of the translation inhibitor 4E-BP1. Genes Dev 15: 2852-2864, 2001.
44. Takahashi S, Shibutani S and Iwata H: Nuclear-targeted 4E-BP1 is dephosphorylated, induces nuclear translocation of eIF4E, and alters mRNA translation. Exp Cell Res 418: 113246, 2022.
45. Xu K, Wang L, Feng W, Feng Y and Shu HK: Phosphatidylinositol-3 kinase-dependent translational regulation of Id1 involves the PPM1G phosphatase. Oncogene 35: 5807-5816, 2016.
46. Sultana S, Zarreen F and Chakraborty S: Insights into the roles of histone chaperones in nucleosome assembly and disassembly in virus infection. Virus Res 297: 198395, 2021.
47. Li S, Edwards G, Radebaugh CA, Luger K and Stargell LA: Spn1 and its dynamic interactions with Spt6, histones and nucleosomes. J Mol Biol 434: 167630, 2022.
48. Lian Y, Hao H, Xu J, Bo T, Liang A and Wang W: The histone chaperone Nrp1 is required for chromatin stability and nuclear division in Tetrahymena thermophila. Epigenetics Chromatin 14: 34, 2021.
49. Kimura H, Takizawa N, Allemand E, Hori T, Iborra FJ, Nozaki N, Muraki M, Hagiwara M, Krainer AR, Fukagawa T and Okawa K: A novel histone exchange factor, protein phosphatase 2Cgamma, mediates the exchange and dephosphorylation of H2A-H2B. J Cell Biol 175: 389-400, 2006.
50. Luo Q, Wang B, Wu Z, Jiang W, Wang Y, Du K, Zhou N, Zheng L, Gan J, Shen WH, *et al*: NAP1-Related Protein 1 (NRP1) has multiple interaction modes for chaperoning histones H2A-H2B. Proc Natl Acad Sci USA 117: 30391-30399, 2020.
51. Farrugia M, Vassallo N and Cauchi RJ: Disruption of survival motor neuron in glia impacts survival but has no effect on neuromuscular function in Drosophila. Neuroscience 491: 32-42, 2022.
52. Riboldi GM, Faravelli I, Kuwajima T, Delestrée N, Dermentzaki G, De Planell-Saguer M, Rinchetti P, Hao LT, Beattie CC, Corti S, *et al*: Sumoylation regulates the assembly and activity of the SMN complex. Nat Commun 12: 5040, 2021.

53. Musawi S, Donnio LM, Zhao Z, Magnani C, Rassinoux P, Binda O, Huang J, Jacquier A, Coudert L, Lomonte P, *et al*: Nucleolar reorganization after cellular stress is orchestrated by SMN shuttling between nuclear compartments. Nat Commun 14: 7384, 2023.
54. Franco-Espin J, Gatius A, Armengol JÁ, Arumugam S, Moradi M, Sendtner M, Calderó J and Tabares L: SMN is physiologically downregulated at wild-type motor nerve terminals but aggregates together with neurofilaments in SMA mouse models. Biomolecules 12: 1524, 2022.
55. Petri S, Grimmler M, Over S, Fischer U and Gruss OJ: Dephosphorylation of survival motor neurons (SMN) by PPM1G/PP2Cgamma governs Cajal body localization and stability of the SMN complex. J Cell Biol 179: 451-465, 2007.
56. Detering NT, Schüning T, Hensel N and Claus P: The phospho-landscape of the survival of motoneuron protein (SMN) protein: Relevance for spinal muscular atrophy (SMA). Cell Mol Life Sci 79: 497, 2022.
57. Husedzinovic A, Neumann B, Reymann J, Draeger-Meurer S, Chari A, Erfle H, Fischer U and Gruss OJ: The catalytically inactive tyrosine phosphatase HD-PTP/PTPN23 is a novel regulator of SMN complex localization. Mol Biol Cell 26: 161-171, 2015.
58. Martinez-Salas E, Embarc-Buh A and Francisco-Velilla R: Emerging roles of Gemin5: From snRNPs assembly to translation control. Int J Mol Sci 21: 3868, 2020.
59. Hu Y, Hou Y, Zhou S, Wang Y, Shen C, Mu L, Su D and Zhang R: Mechanism of assembly of snRNP cores assisted by ICln and the SMN complex in fission yeast. iScience 26: 107604, 2023.
60. Oksenych V and Kainov DE: DNA damage response. Biomolecules 11: 123, 2021.
61. Santivasi WL and Xia F: Ionizing radiation-induced DNA damage, response, and repair. Antioxid Redox Signal 21: 251-259, 2014.
62. Ciccia A and Elledge SJ: The DNA damage response: Making it safe to play with knives. Mol Cell 40: 179-204, 2010.
63. Georgoulis A, Vorgias CE, Chrousos GP and Rogakou EP: Genome instability and γH2AX. Int J Mol Sci 18: 1979, 2017.
64. Beli P, Lukashchuk N, Wagner SA, Weinert BT, Olsen JV, Baskcomb L, Mann M, Jackson SP and Choudhary C: Proteomic investigations reveal a role for RNA processing factor THRAP3 in the DNA damage response. Mol Cell 46: 212-225, 2012.
65. Khoronenkova SV, Dianova II, Ternette N, Kessler BM, Parsons JL and Dianov GL: ATM-dependent downregulation of USP7/HAUSP by PPM1G activates p53 response to DNA damage. Mol Cell 45: 801-813, 2012.
66. Koo N, Sharma AK and Narayan S: Therapeutics targeting p53-MDM2 interaction to induce cancer cell death. Int J Mol Sci 23: 5005, 2022.
67. Williams AB and Schumacher B: p53 in the DNA-damage-repair process. Cold Spring Harb Perspect Med 6: a026070, 2016.
68. Matsuoka S, Ballif BA, Smogorzewska A, McDonald ER III, Hurov KE, Luo J, Bakalarski CE, Zhao Z, Solimini N, Lerenthal Y, *et al*: ATM and ATR substrate analysis reveals extensive protein networks responsive to DNA damage. Science 316: 1160-1166, 2007.
69. Khoronenkova SV and Dianov GL: Regulation of USP7/HAUSP in response to DNA damage: Yet another role for ATM. Cell Cycle 11: 2409-2410, 2012.
70. Chaudhary N and Maddika S: WWP2-WWP1 ubiquitin ligase complex coordinated by PPM1G maintains the balance between cellular p73 and ΔNp73 levels. Mol Cell Biol 34: 3754-3764, 2014.
71. Chen X, Ma J, Wang ZW and Wang Z: The E3 ubiquitin ligases regulate inflammation in cardiovascular diseases. Semin Cell Dev Biol 154: 167-174, 2024.
72. Wang Y, Wu Z, Wang C, Wu N, Wang C, Hu S and Shi J: The role of WWP1 and WWP2 in bone/cartilage development and diseases. Mol Cell Biochem: Jan 22, 2024 (Epub ahead of print).
73. Yoon MK, Ha JH, Lee MS and Chi SW: Structure and apoptotic function of p73. BMB Rep 48: 81-90, 2015.
74. Osterburg C and Dötsch V: Structural diversity of p63 and p73 isoforms. Cell Death Differ 29: 921-937, 2022.
75. Engeland K: Cell cycle regulation: p53-p21-RB signaling. Cell Death Differ 29: 946-960, 2022.
76. Kuganesan N, Dlamini S, Tillekeratne LMV and Taylor WR: Tumor suppressor p53 promotes ferroptosis in oxidative stress conditions independent of modulation of ferroptosis by p21, CDKs, RB, and E2F. J Biol Chem 297: 101365, 2021.
77. Suh EJ, Kim TY and Kim SH: PP2Cgamma-mediated S-phase accumulation induced by the proteasome-dependent degradation of p21(WAF1/CIP1). FEBS Lett 580: 6100-6104, 2006.
78. Desvoyes B and Gutierrez C: Roles of plant retinoblastoma protein: Cell cycle and beyond. EMBO J 39: e105802, 2020.
79. Sanidas I, Morris R, Fella KA, Rumde PH, Boukhali M, Tai EC, Ting DT, Lawrence MS, Haas W and Dyson NJ: A code of mono-phosphorylation modulates the function of RB. Mol Cell 73: 985-1000.e6, 2019.
80. Sharma SS and Pledger WJ: The non-canonical functions of p27(Kip1) in normal and tumor biology. Cell Cycle 15: 1189-1201, 2016.
81. Deshmukh D, Xu J, Yang X, Shimelis H, Fang S and Qiu Y: Regulation of p27 (Kip1) by ubiquitin E3 ligase RNF6. Pharmaceutics 14: 802, 2022.
82. Cassimere EK, Mauvais C and Denicourt C: p27Kip1 is required to mediate a G1 cell cycle arrest downstream of ATM following genotoxic stress. PLoS One 11: e0162806, 2016.
83. Fujita N, Sato S, Katayama K and Tsuruo T: Akt-dependent phosphorylation of p27Kip1 promotes binding to 14-3-3 and cytoplasmic localization. J Biol Chem 277: 28706-28713, 2002.
84. Sun C, Wang G, Wrighton KH, Lin H, Songyang Z, Feng XH and Lin X: Regulation of $p27^{Kip1}$ phosphorylation and G1 cell cycle progression by protein phosphatase PPM1G. Am J Cancer Res 6: 2207-2220, 2016.
85. Sousa B, Pereira J and Paredes J: The crosstalk between cell adhesion and cancer metabolism. Int J Mol Sci 20: 1933, 2019.
86. Sun Y, Zhang J and Ma L: α-Catenin. A tumor suppressor beyond adherens junctions. Cell Cycle 13: 2334-2339, 2014.
87. Lessey LR, Robinson SC, Chaudhary R and Daniel JM: Adherens junction proteins on the move-from the membrane to the nucleus in intestinal diseases. Front Cell Dev Biol 10: 998373, 2022.
88. Duong CN, Brückner R, Schmitt M, Nottebaum AF, Braun LJ, Meyer Zu Brickwedde M, Ipe U, Vom Bruch H, Schöler HR, Trapani G, *et al*: Force-induced changes of α-catenin conformation stabilize vascular junctions independently of vinculin. J Cell Sci 134: jcs259012, 2021.
89. Kumar P, Tathe P, Chaudhary N and Maddika S: PPM1G forms a PPP-type phosphatase holoenzyme with B56δ that maintains adherens junction integrity. EMBO Rep 20: e46965, 2019.
90. Foster WH, Langenbacher A, Gao C, Chen J and Wang Y: Nuclear phosphatase PPM1G in cellular survival and neural development. Dev Dyn 242: 1101-1109, 2013.
91. Chen J, Li J, Sun H, Hu T, Wang Y, Kang G, Cao M and Li X: PPM1G promotes the progression of lung adenocarcinoma by inhibiting p38 activation via dephosphorylation of MEK6. Carcinogenesis 44: 93-104, 2023.
92. Kim EK and Choi EJ: Compromised MAPK signaling in human diseases: An update. Arch Toxicol 89: 867-882, 2015.
93. Di Rocco A, Camero S, Benedetti A, Lozanoska-Ochser B, Megiorni F, Marchese C, Stramucci L, Ciccarelli C, Bouché M, Bossi G, *et al*: Anti-oncogenic and pro-myogenic action of the MKK6/p38/AKT axis induced by targeting MEK/ERK in embryonal rhabdomyosarcoma. Oncol Rep 48: 151, 2022.
94. Martínez-Limón A, Joaquin M, Caballero M, Posas F and de Nadal E: The p38 pathway: From biology to cancer therapy. Int J Mol Sci 21: 1913, 2020.
95. Ming Z, Lim SY and Rizos H: Genetic alterations in the INK4a/ARF locus: Effects on melanoma development and progression. Biomolecules 10: 1447, 2020.
96. Hyder U, McCann JL, Wang J, Fung V, Bayo J and D'Orso I: The ARF tumor suppressor targets PPM1G/PP2Cγ to counteract NF-κB transcription tuning cell survival and the inflammatory response. Proc Natl Acad Sci USA 117: 32594-32605, 2020.
97. Roschger C and Cabrele C: The Id-protein family in developmental and cancer-associated pathways. Cell Commun Signal 15: 7, 2017.
98. Xiong DL, Li Q, Wang H, Jin WL, Fan XM and Ma YY: High expression of PPM1G is associated with the progression and poor prognosis of hepatocellular carcinoma. Cancer Biomark 34: 13-22, 2022.
99. Hu W, Ma SL, Qiong L, Du Y, Gong LP, Pan YH, Sun LP, Wen JY, Chen JN, Guan XY and Shao CK: PPM1G promotes cell proliferation via modulating mutant GOF p53 protein expression in hepatocellular carcinoma. iScience 27: 109116, 2024.
100. Liu X, Zeng J, Li H, Li F, Jiang B, Zhao M, Liu Z, Li R and Ma T: A risk model based on sorafenib-response target genes predicts the prognosis of patients with HCC. J Oncol 2022: 7257738, 2022.
101. Li W, Liu J, Zhang D, Gu L and Zhao H: The prognostic significance and potential mechanism of ferroptosis-related genes in hepatocellular carcinoma. Front Genet 13: 844624, 2022.

102. Yang H, Huo J and Li X: Identification and validation of a five-gene prognostic signature for hepatocellular carcinoma. World J Surg Oncol 19: 90, 2021.
103. Li W, Lu J, Ma Z, Zhao J and Liu J: An integrated model based on a six-gene signature predicts overall survival in patients with hepatocellular carcinoma. Front Genet 10: 1323, 2020.
104. Zhang BH, Yang J, Jiang L, Lyu T, Kong LX, Tan YF, Li B, Zhu YF, Xi AY, Xu X, *et al*: Development and validation of a 14-gene signature for prognosis prediction in hepatocellular carcinoma. Genomics 112: 2763-2771, 2020.
105. Jia R and Zheng ZM: Oncogenic SRSF3 in health and diseases. Int J Biol Sci 19: 3057-3076, 2023.
106. Sen S, Langiewicz M, Jumaa H and Webster NJG: Deletion of serine/arginine-rich splicing factor 3 in hepatocytes predisposes to hepatocellular carcinoma in mice. Hepatology 61: 171-183, 2015.
107. Wen C, Tian Z, Li L, Chen T, Chen H, Dai J, Liang Z, Ma S and Liu X: SRSF3 and HNRNPH1 regulate radiation-induced alternative splicing of protein arginine methyltransferase 5 in hepatocellular carcinoma. Int J Mol Sci 23: 14832, 2022.
108. Hong B, van den Heuvel AP, Prabhu VV, Zhang S and El-Deiry WS: Targeting tumor suppressor p53 for cancer therapy: Strategies, challenges and opportunities. Curr Drug Targets 15: 80-89, 2014.
109. Zhang C, Liu J, Xu D, Zhang T, Hu W and Feng Z: Gain-of-function mutant p53 in cancer progression and therapy. J Mol Cell Biol 12: 674-687, 2020.
110. Zhao M, Wang T, Gleber-Netto FO, Chen Z, McGrail DJ, Gomez JA, Ju W, Gadhikar MA, Ma W, Shen L, *et al*: Mutant p53 gains oncogenic functions through a chromosomal instability-induced cytosolic DNA response. Nat Commun 15: 180, 2024.
111. Roehlen N, Crouchet E and Baumert TF: Liver fibrosis: Mechanistic concepts and therapeutic perspectives. Cells 9: 875, 2020.
112. Ge MX, Liu HT, Zhang N, Niu WX, Lu ZN, Bao YY, Huang R, Yu DK, Shao RG and He HW: Costunolide represses hepatic fibrosis through WW domain-containing protein 2-mediated Notch3 degradation. Br J Pharmacol 177: 372-387, 2020.
113. Reig M, Forner A, Rimola J, Ferrer-Fàbrega J, Burrel M, Garcia-Criado Á, Kelley RK, Galle PR, Mazzaferro V, Salem R, *et al*: BCLC strategy for prognosis prediction and treatment recommendation: The 2022 update. J Hepatol 76: 681-693, 2022.
114. Guan Y, Yao W, Yi K, Zheng C, Lv S, Tao Y, Hei Z and Li M: Nanotheranostics for the management of hepatic ischemia-reperfusion injury. Small 17: e2007727, 2021.
115. Peng D, Huang Z, Yang H, Luo Y and Wu Z: PPM1G regulates hepatic ischemia/reperfusion injury through STING-mediated inflammatory pathways in macrophages. Immun Inflamm Dis 12: e1189, 2024.
116. Yu K, Tian H and Deng H: PPM1G restricts innate immune signaling mediated by STING and MAVS and is hijacked by KSHV for immune evasion. Sci Adv 6: eabd0276, 2020.
117. Ho SY, Chang CM, Liao HN, Chou WH, Guo CL, Yen Y, Nakamura Y and Chang WC: Current trends in neoantigen-based cancer vaccines. Pharmaceuticals (Basel) 16: 392, 2023.
118. Fu J, Chen F, Lin Y, Gao J, Chen A and Yang J: Discovery and characterization of tumor antigens in hepatocellular carcinoma for mRNA vaccine development. J Cancer Res Clin Oncol 149: 4047-4061, 2023.
119. Wang B, Pei J, Xu S, Liu J and Yu J: Recent advances in mRNA cancer vaccines: Meeting challenges and embracing opportunities. Front Immunol 14: 1246682, 2023.
120. Lu TL, Li CL, Gong YQ, Hou FT and Chen CW: Identification of tumor antigens and immune subtypes of hepatocellular carcinoma for mRNA vaccine development. World J Gastrointest Oncol 15: 1717-1738, 2023.