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REVIEW

Stress-activated kinases as therapeutic targets in pancreatic cancer

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Abstract

Pancreatic cancer is a dismal disease with high incidence and poor survival rates. With the aim to improve overall survival of pancreatic cancer patients, new therapeutic approaches are urgently needed. Protein kinases are key regulatory players in basically all stages of development, maintaining physiologic functions but also being involved in pathogenic processes. c-Jun N-terminal kinases (JNK) and p38 kinases, representatives of the mitogen-activated protein kinases, as well as the casein kinase 1 (CK1) family of protein kinases are important mediators of adequate response to cellular stress following inflammatory and metabolic stressors, DNA damage, and others. In their physiologic roles, they are responsible for the regulation of cell cycle progression, cell proliferation and differentiation, and apoptosis. Dysregulation of the underlying pathways consequently has been identified in various cancer types, including pancreatic cancer. Pharmacological targeting of those pathways has been the field of interest for several years. While success in earlier studies was limited due to lacking specificity and off-target effects, more recent improvements in small molecule inhibitor design against stress-activated protein kinases and their use in combination therapies have shown promising *in vitro* results. Consequently, targeting of JNK, p38, and CK1 protein kinase family members may actually be of particular interest in the field of precision medicine in patients with highly deregulated kinase pathways related to these kinases. However, further studies are warranted, especially involving *in vivo* investigation and clinical trials, in order to advance inhibition of stress-activated kinases to the field of translational medicine.

Key Words: Pancreatic cancer; Stress-activated protein kinases; Mitogen-activated protein kinases; c-Jun N-terminal kinases; Casein kinase 1; Small molecule inhibitor

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Core Tip: Since pancreatic cancer patients are generally confronted with poor prognosis, optimized therapeutic strategies are urgently needed. To establish new treatment options, efforts in drug development have increasingly focused on targeting protein kinases. In the cellular response to various stress signals, c-Jun N-terminal kinases (JNK) and p38 kinases as well as members of the casein kinase 1 (CK1) family are of special interest. Concentrating on pancreatic carcinoma in this review, we summarize the key roles of JNK, p38, and CK1 and provide an overview of recent achievements in the development of small molecule kinase inhibitors against these kinases.

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INTRODUCTION

Pancreatic cancer is a severe disease, with overall 5-year survival rates less than 10% and only very little improvement over the last decades[1]. It is currently the fourth most common cause of cancer-related deaths, and it is expected by 2030 that pancreatic cancer will have surpassed colon and breast/prostate cancer to move up to second rank of cancer-related deaths[2]. Contributing to the immense challenge of treating pancreatic cancer, dysregulation of multiple signaling pathways can frequently be detected. With the genetic hallmark mutation of *KRAS* in over 90% of all pancreatic cancer patients, the high relevance of kinase-driven pathways is underlined[3].

So far, classic chemotherapeutic agents have only shown moderate success in prolonging overall survival of patients suffering from pancreatic ductal adenocarcinoma (PDAC). However, more and more personalized therapy concepts are becoming promising options, especially the use of small molecule inhibitors specifically targeting newly identified drug targets, such as deregulated protein kinases. Of special interest are kinases activated in cellular stress situations, like mitogen-activated protein kinases (MAPKs) and members of the casein kinase 1 (CK1) family, which phosphorylate signal integration molecules like p53 and β-catenin finally resulting in activation of processes leading to cell cycle arrest or apoptosis.

MAPKs

MAPKs are key players in transducing extracellular stimuli into intracellular signaling cascades and therefore represent interesting drug targets. Multiple isoforms have been identified, which can be clustered into six groups of MAPKs. The most prominent of those are the extracellular-regulated kinases 1 and 2 (ERK1/2), the c-Jun N-terminal kinases 1, 2, and 3 (JNK1/2/3), and the p38 kinases α, β, γ, and δ[4,5]. As a response to various stimuli such as growth factors, cytokines, and environmental stress, MAPK-triggered phosphorylation of their target transcription factors (TFs) marks the endpoint of an intracellular kinase cascade. This cascade consists of ligands binding to their cell membrane receptors, recruitment of GTPase (*e.g.*, RAS) to the plasma membrane, and activation of MAPK kinase kinases (MKKKs or MAPKKKs, *e.g.*, RAF) as well as MAPK kinases (MKKs or MAPKKs, *e.g.*, MEK1/2)[5-7]. ERK1/2 belong to the best-studied kinases among MAPKs. Their relevance for pancreatic cancer has been well documented, especially as ERKs exert their functions downstream of mutant KRAS[8,9]. JNK and p38 can be grouped together as stress-activated protein kinases (SAPKs), as their pathways are regularly activated by environmental stressors, like nutrient deprivation, inflammatory cytokines, or ultraviolet irradiation[5,10].

CK1

A remarkable association with tumorigenesis and tumor progression has also been demonstrated for the CK1 family of protein kinases. Being among the first kinases described in history, involvement of CK1 isoforms in several essential signal transduction pathways has been reported within the last decades. As a key cascade in developmental processes, the (canonical) Wnt/β-catenin signaling pathway can also be involved in promoting cell proliferation through activation of oncogenes like c-myc

and cyclin D1[11,12]. All human CK1 isoforms were identified to fulfill negative as well as positive regulatory functions in canonical Wnt signaling, thereby either acting as tumor suppressors or contributing to Wnt-induced oncogenic processes[13-15]. CK1δ and ε might furthermore promote canonical instead of non-canonical Wnt signaling, consequently resulting in reduced JNK-mediated Wnt signaling and apoptosis[16,17]. In addition, apoptosis mediated by Fas can also be down-regulated by CK1δ- and ε-mediated stabilization of Bid[18]. Apart from signaling associated with proliferation, differentiation, and apoptosis, CK1 is also involved in further mechanisms of the cellular stress response, including functions in immune response and inflammation, regulation of microtubule dynamic processes, autophagy, and DNA damage-related signal transduction[19-21]. Especially well documented is the regulatory function of CK1 isoforms in p53-mediated signal transduction with CK1δ even forming an autoregulatory feedback loop with p53[19].

Cancer itself already forms a stressful environment on the tumor cells, induced by hypoxia and nutrient deprivation as well as metabolic and replication stress. Additionally, cancer cells face genotoxic stress exerted by chemo- and radiotherapies. In this regard, pancreatic cancer is no exception since tumors of the pancreas are known for their dense stroma with impaired vasculature and the association with metabolic stressors, like diabetic conditions. This review aims to elucidate the role of the stress-activated kinases: JNK and p38 but also CK1 in the pathogenesis of pancreatic cancer and their potential as therapeutic targets.

SAPKS: JNK AND P38

The first description of JNK as a 54 kDa protein kinase activated upon peritoneal cycloheximide injection into rats dates back to 1990[22]. So far, three different JNK isoforms have been identified in the human genome, including JNK1 (MAPK8), JNK2 (MAPK9), and JNK3 (MAPK10), whereas expression of JNK3 is mainly restricted to brain, heart, and testes[23]. Alternative splicing results in generation of at least 10 different JNK isoforms with molecular weights ranging from 46 kDa to 55 kDa[24]. Activation of JNKs is dependent on phosphorylation of threonine and tyrosine residues by upstream kinases. For JNK1, phosphorylation of Thr^{180} and Tyr^{182} within its kinase subdomain VIII has been demonstrated to be essential[25]. In the activation process of JNK1, the upstream kinases MKK4 and MKK7 both fulfill non-redundant functions, with MKK4 preferably phosphorylating Tyr residues while MKK7 favors Thr residues. Phosphorylation of Thr^{180} is sufficient for JNK1 activation, however, dual phosphorylation by both kinases is required for full JNK activation[26,27]. Further upstream of the signaling cascade, a large variety of at least 14 MKKKs can activate MKK4 and MKK7[23,28].

Merging the influence of multiple upstream kinases into fewer effector kinases enables the cells to respond to a variety of stimuli, like growth factors and cytokines, reactive oxygen species, physical interactions with other cells and the extracellular matrix, as well as cellular stressors[29]. The variety of different stimuli requires multiple cell membrane receptors engaging into the JNK pathway. These include G-protein coupled receptors, Wnt-receptors, transforming growth factor-β receptors, and tumor necrosis factor-α (TNF-α) receptors[30]. Signal transfer within the SAPK pathway is generally orchestrated by docking motifs for upstream kinases and downstream substrates, as well as scaffold proteins. Those scaffold proteins express docking sites for MKKKs, MKKs, and MAPKs and play an important role in the correct stimulus response through the kinase cascade[29,31].

Even more diverse than the upstream mediators are the possible JNK substrates. As all MAPKs, JNK is a proline-targeted serine/threonine kinase, thus preferably phosphorylating Ser-Pro as well as Thr-Pro motifs[4,32]. So far, the list of JNK substrates includes more than 100 targets, among them TFs like c-Jun, p53, c-myc, and β-catenin, microtubule-associated proteins, components of focal-adhesion-complex and cell-to-cell-adhesion, as well as apoptosis-regulating proteins like Bcl-2 and Bax [30]. Figure 1 shows an overview of MAPK-related cellular functions.

The first p38 MAPK was discovered in 1994, and today four isoforms (p38α, β, γ, and δ, corresponding to MAPK14, 11, 12 and 13) are known[33,34]. While p38α is ubiquitously expressed, the other isoforms show differential tissue distribution, with p38β being mainly expressed in the brain, p38γ in skeletal muscle, and p38δ in endocrine glands[35]. Dual phosphorylation of Thr^{180} and Tyr^{182} in a Thr-Gly-Tyr motif are required for full p38 kinase activation[36]. MKK3 and MKK6 specifically activate p38, but while MKK6 can activate all isoforms, MKK3 is unable to phosphorylate p38β

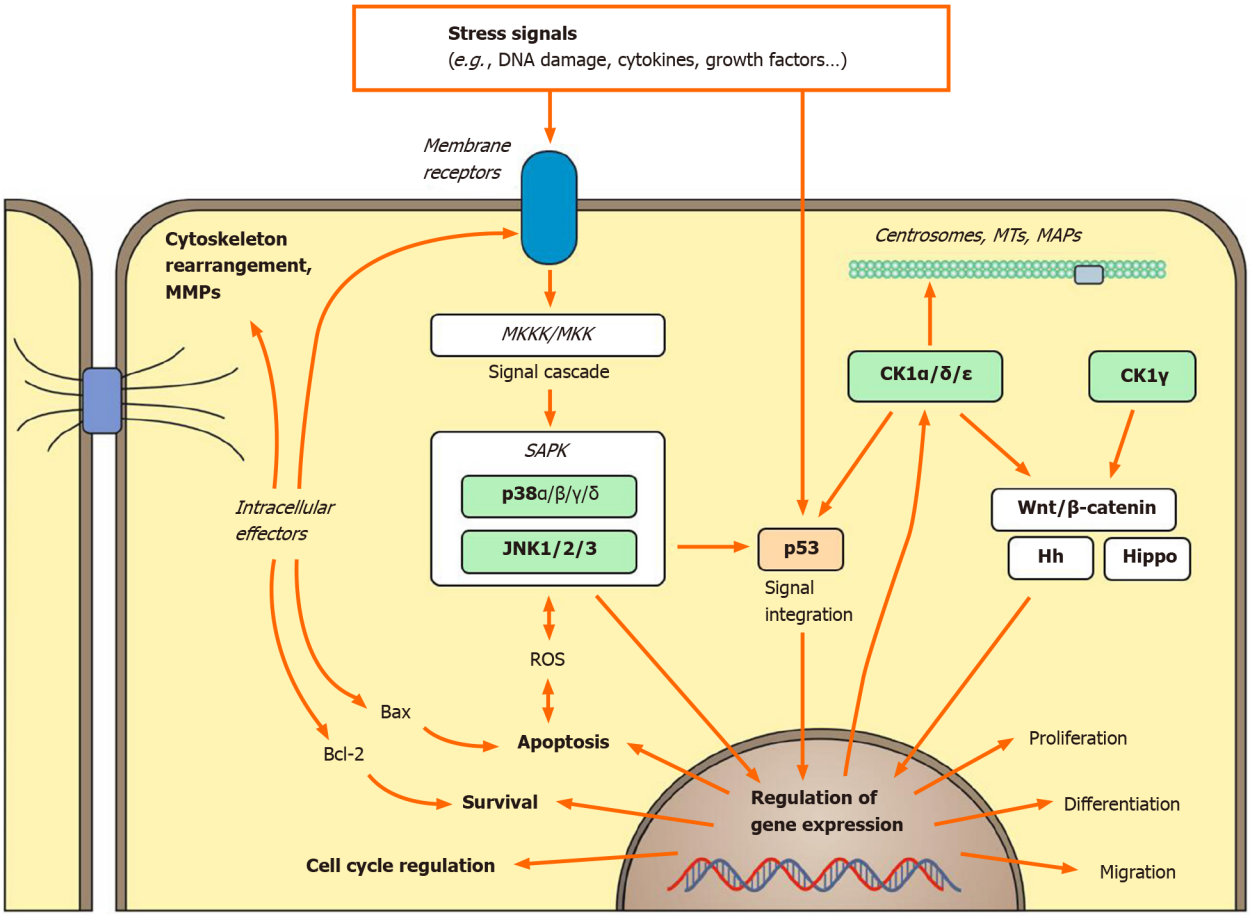

Figure 1 Signal integration mediated by stress-activated protein kinases. The stress-activated protein kinases (SAPKs) c-Jun N-terminal kinase (JNK) and p38 as well as the protein kinases of the casein kinase 1 (CK1) family are activated in response to endogenous and exogenous stress stimuli. Therefore, JNK and p38 are either activated directly or through upstream signaling cascades [mitogen-associated protein kinase kinase kinases (MKKKs), mitogen-associated protein kinase kinases (MKKs)] and subsequently exercise their functions through intracellular signal integrating effectors such as the transcription factors JNK, p53, c-myc, or β-catenin. In response to certain stress stimuli, the kinases of the CK1 family also take key functions in regulatory effects mediated *via* p53 or through various signal transduction pathways like the Wnt/β-catenin, Hedgehog (Hh), or Hippo pathway. Finally, cellular stress response including regulation of proliferation, differentiation, migration, cell cycle progression, survival, and apoptosis is initiated by modulation of gene expression. MAP: Microtubule-associated protein; MMP: matrix metalloproteinase; MT: Microtubule; ROS: Reactive oxygen species.

[37,38]. MKK4 can also contribute to p38 activation[39,40]. Multiple MKKKs contribute to the activation of MKK3 and MKK6, among which some are shared with the JNK pathway. By engaging specific MKKKs in response to defined stimuli, cells are enabled to elicit the correct stress response[35]. In T cells, a kinase cascade-independent pathway of p38α activation has also been described[41]. Similar to JNK, there are many p38-specific substrates, ranging from TFs to other protein kinases and apoptosis-regulating proteins[35].

Relevant SAPK-related pathways in cancer

The involvement of JNK and p38 in the pathogenesis of cancer has been studied extensively. Their role has been debated controversially since both can exhibit pro- as well as anti-tumorigenic functions[42,43]. For both kinases, the cellular effects provoked by JNK and p38 depend on the type, strength, and duration of the stimulus [44,45]. However, the influence of SAPKs on cancer development and progression is apparent as major cancer characteristics like cell proliferation, migration, and apoptosis are influenced by p38 and JNK. Pathways associated with these characteristics will now be discussed in general and specifically in pancreatic cancer.

A primary example of differential regulation in SAPK-related pathways is the interaction of JNKs and c-Jun. In non-stimulated cells, JNK2 activity leads to degradation of c-Jun, while after stimulation, JNK1 phosphorylates and stabilizes c-Jun. Consequently, knockdown of JNK1 decreased fibroblast proliferation through reduced activity of Activator protein-1 (AP-1), a TF of which phospho-c-Jun is a vital component[46,47]. Phosphorylation of c-Jun has also been identified as a critical step in RAS-induced tumorigenesis. Oncogenic RAS uses the same phosphorylation sites as

JNK on c-Jun and promotes transformation of rat embryonic fibroblasts, while *c-Jun*^{null} cells are resistant to RAS-induced transformation[48,49]. However, the role of JNK in this process is under dispute. In *TP53*$^{-/-}$ mouse embryonic fibroblasts, dual knockout of JNK1 and 2 reduced *KRAS*^{G12D}-induced transformation and suppressed *in vivo* growth. Furthermore, *KRAS*^{G12D}-induced lung tumor formation in mice was similarly reduced by JNK knockout[50]. This effect was in part attributed to JNK2, as only JNK2- but not JNK1-deprived mouse embryonic fibroblasts resisted RAS-induced transformation, although increased levels of AP-1 and phospho-c-Jun were observed[51]. However, not all transforming effects of RAS seem to be controlled by JNK, and in contrast to the before-mentioned studies, loss of JNK in RAS mutant cells may also contribute to enhanced tumorigenesis through apoptosis regulation[52]. A possible explanation for these findings is JNK-controlled cell cycle progression, as fibroblasts with knockdown of JNK2 show faster G1/S progression, while their JNK1-deprived counterparts show an opposing phenotype[46].

Besides proliferation, pro- and anti-apoptotic signals are also mediated by JNK in dependency of the stimulus. JNK regulates the expression of Bcl-2 family members and thereby influences apoptosis mediated *via* the mitochondrial pathway[44]. In TNF-mediated apoptosis, at early time-points, JNK activation triggers pro-survival pathways, while functioning JNK signaling is required for TNF-mediated apoptosis under persistent stimulation[53]. In murine cancer models, JNK1 was shown to promote chemically induced liver cancer, a finding that was also confirmed in human hepatocellular carcinoma[54,55]. On the other hand, knockdown of JNK1 rendered mice more susceptible to chemically induced skin tumors, while knockdown of JNK2 exerted an opposite effect[56].

The influence of p38 on oncogenesis is generally thought to be tumor suppressive, but protumorigenic functions like promotion of invasiveness has also been reported [44]. p38α controls proliferation through regulation of cell cycle progression at the G1/S and G2/M phases[57]. p38 can inhibit G1/S progression, *e.g.*, through downregulation of cyclin D1[40,58] or by phosphorylation of p53 and retinoblastoma protein [59-61]. Alternatively, p38 can also promote cell cycle progression through the induction of cyclin A or interference with the retinoblastoma protein pathway[62,63]. p38-mediated phosphorylation of p53 also activates the G2/M checkpoint in response to DNA double-strand breaks. Ironically, this also offers a survival benefit for tumor cells and increases therapy resistance, as DNA damaging drugs become less effective with functional p38 signaling[64-66]. Other tumor-promoting roles include formation of a pro-invasive phenotype through induction of matrix metalloproteinases and tumor cell dormancy, enabling metastatic relapse[42].

p38-related implication for involvement of the SAPK pathway in pancreatic cancer

The influence of the SAPK pathway on pancreatic cancer has been studied on patient samples as well as genetically engineered cell lines and mouse models, while patient cohorts are especially helpful to study population risk factors. Handra-Luca *et al*[67] offered an immunohistochemical analysis of the MAPK pathway in 99 surgically resected pancreatic cancer specimens. While high immunoreactivity for ERK1/2 was consistently associated with a worse prognosis, high expression levels of p38 could be associated with shorter recurrence-free survival in patients without adjuvant treatment. Strong staining for MKK4 was associated with increased proliferation[67]. Contrarily hereto, phosphorylated p38 with activated downstream TFs was identified as a favorable biomarker after surgical resection and associated with reduced number of lymph node metastases. Labeling of phospho-p38 showed no changes through the different cancer stages. Furthermore, pharmacologically inhibiting p38 *in vitro* and *in vivo* resulted in enhanced JNK signaling and enhanced cell growth[68]. Phospho-p38 was reported to increase during tumor progression, which is consistent with reports claiming that p38 has tumor suppressive functions during early carcinogenesis but switches towards a tumor promoting phenotype later on[69]. In our own previous work, we were able to dissect the isoform-specific functions of p38 in pancreatic cancer by genetically targeting p38 isoforms α and β. We confirmed an *in vivo* tumor suppressive phenotype of p38α but also showed a pro-invasive function. Additionally, we showed a tumor suppressive role of p38β, opposing p38α[70]. Interestingly, oncogenic KRAS induced activation of p38 and phenotypically increased invasion[71, 72].

JNK-related implication for involvement of the SAPK pathway in pancreatic cancer

Increased phospho-JNK staining was observed in pancreatic cancer tissues compared to normal controls[73] and increased phospho-JNK1 staining was determined as an independent predictor of peritoneal spread[74]. Furthermore, serum auto-antibodies

against JNK2 were identified as potential biomarker in pancreatic cancer patients[75]. By isoform-specific knockdown of JNK1 and 2 in MiaPaCa-2 and Panc-1 cells, a tumor promoting role could be attributed to JNK1, while JNK2 seems to exert suppressive functions in pancreatic cancer[76]. Upstream kinases of JNK have also been studied in pancreatic cancer, but their distinct role in tumor formation and progression remains elusive. MEKK1, as a representative of JNK-activating MKKKs, was shown to contribute to pancreatic cancer cell survival. However, unlike in other cancer cell lines, JNK signaling was not affected by knockout of MEKK1 in the PDAC cell line Panc-1 [77]. MKK4, a direct upstream kinase of JNK, was expressed in the majority of resected specimens, while expression levels were reduced in matched metastatic samples. As further hints for a potential tumor suppressive role, patients with loss of MKK4 were associated with shorter survival, and pancreatic cancer cell lines frequently showed loss-of-heterozygosity for MKK4[78,79]. However, ectopic expression of MKK4 stimulated proliferation and migration of ASPC-1 and BxPC3 cells[80].

Both kinases also act in pancreatic cancer outside neoplastic cells. *Ptf1a*$^{Cre/+}$; *Kras*$^{G12D/+}$; *JNK1*$^{-/-}$ mice showed significantly smaller tumors than their *JNK1*$^{+/-}$ counterparts. Tumors induced by transplantation of murine PDAC cells were larger in wild-type mice than in *JNK1*$^{-/-}$ mice lacking JNK signaling in stromal and immune cells. Interestingly, mice heterozygous for JNK1 showed less infiltrating $CD8^+$ T cells, possibly due to JNK-mediated downregulation of chemokine secretion of tumor-associated fibroblasts[81]. On the other hand, alternative activation of p38 through the T cell receptor in $CD4^+$ T cells resulted in more aggressive disease through secretion of pro-inflammatory cytokines like interleukin-17 and TNF-α[82].

SAPK inhibitors and their role for the treatment of pancreatic cancer

Due to the involvement of SAPKs within a variety of cellular processes and diseases, multiple researchers and the pharmaceutical industry have focused on identifying pharmacological inhibitors.

Generally, JNK small molecule inhibitors can be grouped into adenosine triphosphate (ATP)-competitive and non-ATP-competitive inhibitors. ATP-competitive inhibitors represent the majority of compounds, and most of them act as pan-JNK inhibitors, as the ATP-binding pocket is highly conserved among all three isoforms [83]. Non-ATP-competitive inhibitors target the interaction of JNK with upstream and downstream targets as well as scaffolding proteins. p38 inhibitors can also be grouped by their way of action. Similar to JNK inhibitors, many p38 inhibitors act as ATP-competitive inhibitors, either binding to the active (type 1 inhibitors) or inactive conformation of p38 (type 2 inhibitors)[84]. So far, p38 as well as JNK have been targeted in therapeutic intention for multiple pathologic conditions, like neurodegenerative diseases (*e.g.*, Alzheimer's and Parkinson's disease) and inflammatory diseases (*e.g.*, rheumatoid arthritis and inflammatory bowel disease)[85-87]. Consequently, pharmacological inhibition of both kinases has also been tested in order to explore new therapeutic strategies for the treatment of neoplastic diseases. Table 1 summarizes selective SAPK inhibitors studied in the context of cancer.

Strikingly, most studies report an antitumor effect of pharmacological SAPK inhibition. However, it needs to be noted that all inhibitors were pan-JNK inhibitors and p38α or pan-p38 inhibitors, respectively. As mentioned above, we previously reported on marked isoform-specific differences. By using genetic pathway disruption, isoform-specific tumor suppressive functions of JNK2 and p38β were detected and consequently, targeting of these isoforms might increase the risk of failure in clinical studies[76]. This effect could not be observed in our study, as pharmacological inhibition of JNK also reduced cell growth in various cell lines. Up to now, clinical studies using p38 inhibitors failed or only showed moderate success. Only one JNK inhibitor (CC-401) has been clinically evaluated for the treatment of cancer (NCT00126893), but this study has been discontinued[88,89].

In an early study, Ding *et al*[90] showed that the p38 inhibitor SB203580 [half maximal inhibitory concentration (IC_{50}) = 34 nmol/L] increases the number of Panc-1 cells in S phase as well as their proliferation. Decreased p38 activity was confirmed by detection of reduced levels of phospho-activating transcription factor. However, the same study also revealed increased phosphorylation levels of ERK1/2 and JNK[90]. Increased activation of ERK1/2, possibly as a compensation mechanism or off-target effect under SB203580 treatment, has also been shown in other reports[91,92]. Therefore, it remains unclear if the observed increased proliferation is a consequence of the loss of p38-dependent tumor suppressive actions or rather of increased ERK1/2 signaling. More recently, off-target effects of SB203580 and the closely related compound SB202190 were also described by Shanware and colleagues[93], reporting on cellular effects interestingly caused by off-target inhibition of CK1. Similar to the

Table 1 Small molecule inhibitors of stress-activated protein kinases tested for treatment effects on pancreatic carcinoma cells *in vitro* and *in vivo*

Inhibitor	IC_{50} (μmol/L)	Observed effects in cell culture and *in vivo* data	Ref.
JNK inhibitor II(SP600125)	0.040 (JNK1); 0.040 (JNK2); 0.090 (JNK3)	Antitumor effects in cancer cell lines of thyroid, stomach, lung, colon, pancreas, and brain	[104, 185-189]
JNK inhibitor XVI(JNK-IN-8)	0.005 (JNK1); 0.019 (JNK2); 0.980 (JNK3)	Covalent binding to JNK inactivates kinase function; Sensitizes pancreatic cancer cells and triple negative breast cancer cells to 5-FU/FOLFOX and triple negative breast cancer cells to lapatinib treatment	[190-192]
Bentamapimod(AS602801)	0.080 (JNK1); 0.090 (JNK2); 0.230 (JNK3)	Cytotoxic effects observed on cancer stem cells derived from pancreatic cancer, non-small cell lung cancer, ovarian cancer, and glioblastoma	[103, 193]
SB203580	0.034 (p38)	Synergistic effects observed in combination with cisplatin *in vitro* and *in vivo*; Inhibition of gemcitabine-induced apoptosis in combination therapy (tested on PK-1 and PCI-43 PDAC cell lines); $IC_{50(p38)}$ = 0.08-0.20 μmol/L *in vivo*)	[194-198]
SB202190	0.050 (p38α); 0.100 (p38β2); 0.600 (CK1)	Inhibition of gemcitabine-induced apoptosis in combination therapy (tested on PK-1 and PCI-43 PDAC cell lines); Inhibits resistance of colon cancer cell lines towards irinotecan	[93,197, 199,200]
SB239063	0.044 (p38α and β)	Dose-dependent growth inhibition observed in three pancreatic cancer cell lines	[68,201]

CK1: Casein kinase 1; EC_{50}: Half maximal effective concentration; 5-FU: 5-fluorouracil; IC_{50}: Half maximal inhibitory concentration; JNK1/2/3: c-Jun N-terminal kinases 1, 2, and 3.

above mentioned studies, Zhong *et al*[68] reported on the growth enhancing effects not only for SB203580 but also for SB202190 and SB239063 on three different pancreatic cancer cell lines. Interestingly, while environmental stressors (hypoxia and reduced serum levels) led to reduced proliferation in PDAC cell lines, p38 inhibition abolished these effects. Again, increased phospho-JNK levels after p38 inhibition were reported, and JNK inhibition through SP600125 abolished the effects of p38 inhibition *in vitro*. The pan-JNK inhibitor SP600125 ($IC_{50(JNK1)}$ = 40 nmol/L; $IC_{50(JNK2)}$ = 40 nmol/L; $IC_{50(JNK3)}$ = 90 nmol/L) also reduced the *in vivo* growth of cell lines with high phospho-p38 Levels [68]. The crosstalk of p38 and JNK has been described in various contexts previously. Although there is evidence for a synergistic role of both SAPKs in activation of downstream targets[94], an opposing function of both has also been well documented [95-97]. Possible regulation mechanisms of JNK through p38 include upstream MKKKs (MLK3, TAK1) as well as nuclear factor-κB[44]. Consequently, targeting single MAPKs is highly challenging. An alternative approach to using inhibitors could be selectively activating pathways of interest. The small molecule triptonide was shown to selectively activate the MEKK4-MKK4-p38 pathway without significantly altering phosphorylation levels of JNK and ERK1/2. This resulted in dose-dependent growth reduction of six pancreatic cancer cell lines as well as *in vivo* xenografts by inducing G2/M arrest and reduced expression of cyclin-dependent kinase 3[98].

In contrast to those studies showing an overall growth restraining effect of p38 in pancreatic cancer, Yang *et al*[99] performed a screen of p38α expression in various cancer samples of The Cancer Genome Atlas database and identified an overexpression of p38α in PDAC samples. The same study also reported enhanced phospho-p38 labeling in PDAC tissues compared to adjacent normal tissue and mostly attributed phospho-p38 labeling to cancer cells. When treating Pan02 cells with SB203580 or the p38α- and β-specific inhibitor LY2228820, Yang *et al*[99] reported growth-restricting effects. However, it needs to be noted that the used inhibitor concentrations were relatively high and potential off-target effects cannot be excluded. Finally, in order to address the issue of lacking sensitivity, possible binding pockets in p38, enabling the design of more selective inhibitor compounds in future, were identified by *in silico* modeling[99].

Previous studies indirectly suggested a growth-promoting effect of JNKs on pancreatic cancer. Takahashi *et al*[73] observed growth inhibition *in vitro* and *in vivo* after treatment with SP600125. This was associated with G1 arrest and downregulation of cyclin D1 *in vitro*. In genetically-engineered mouse models (*Ptf1a*$^{cre/+}$, *LSL-Kras*$^{G12D/+}$, and *Tgfbr2*$^{flox/flox}$) SP600125 reduced neoangiogenesis and expression levels of CD44 in PDAC cells[73]. Together with CD133, CD44 is considered as a potential marker for cancer stem cells (CSCs) or CSC-like cells (CSCLCs)[100,101]. Increased levels of phospho-JNK was also shown in CSCLCs of pancreatic cancer and other human malignancies[102]. Inhibition of JNK by the pan-JNK inhibitors SP600125 or AS602801 as well as genetic targeting of JNK1 and 2 *via* small interfering RNA-mediated knockdown reduced levels of $CD133^+$ cells in an isolated subpopulation of pancreatic

cancer-derived CSCLCs and abolished their self-renewal capacity *in vitro* and *in vivo* [102,103]. Besides growth suppression and interference with CSCs, induction of cellular differentiation can be another mechanism of JNK inhibition-mediated tumor suppression[104].

In clinical practice, SAPK inhibition will rather be used for combination therapy approaches instead of single-agent therapy. Therefore, the interference of SAPK inhibitors with standard of care chemotherapeutics is highly relevant. In addition to single treatment of CSCLCs, the group of Suzuki *et al*[105] also investigated the effects of JNK inhibition in combination with gemcitabine and 5-fluorouracil (5-FU). While CSCLCs expectedly were more resistant to these agents, pretreatment with SP600125 had a synergistic effect in combination with gemcitabine and 5-FU in a reactive oxygen species-based way of action[105]. The observed synergistic effect can furthermore be explained by JNK-mediated effects on multidrug resistance. Multidrug resistance is not only a hallmark of CSCs but of cancer cells in general, and multidrug transporters like P-glycoprotein reduce intracellular drug levels[106]. In this context, high JNK levels were shown to decrease P-glycoprotein levels in pancreatic and gastric cancer, thereby increasing intracellular drug concentrations as well as drug sensitivity[107].

The interplay of the p38 pathway and gemcitabine treatment has been well studied in pancreatic cancer cells. Apoptosis mediated through gemcitabine was consistently associated with p38 activation as well as caspase-dependent cleavage of poly (ADP-ribose) polymerase and heat shock protein 27 phosphorylation. Inhibition of p38 by SB203580 reversed these effects. Similarly, inhibition of MAPK-activated protein kinase 2, a downstream target of p38, abolished gemcitabine-mediated apoptosis in pancreatic cancer. However, combination of p38 inhibitors with mitomycin C showed synergistic effects[108].

PROTEIN KINASE CK1 FAMILY

In 1954 for the first time, an enzyme was isolated from liver tissue, which was able to phosphorylate the milk protein casein[109]. Fifteen years later, two distinct protein kinases with the ability to phosphorylate casein (at least *in vitro*) were described and termed casein kinase 1 (CK1) and casein kinase 2 (CK2), meanwhile renamed protein kinases CK1 and CK2[110]. Despite their common nomenclature and the ability to phosphorylate casein, protein kinases CK1 and CK2 are highly different with respect to their classification and cellular functions. While CK2 belongs to the CMGC [containing cyclin-dependent kinase, MAPK, glycogen synthase kinase 3 (GSK3), and cdc2-like kinase families] group of the human kinome, CK1 forms an independent family of protein kinases[111].

In the human genome, six CK1 isoforms are encoded (α, γ1, γ2, γ3, δ, and ε), and several splice variants can originate from post-transcriptional processing. While all CK1 isoforms are highly conserved within the kinase domain, the sequences of the N- and C-terminal noncatalytic domains can be quite variable[20,112]. Although the protein kinases of the CK1 family are generally considered to be constitutively active, several regulatory mechanisms have been described. Expression and/or activity levels of CK1 isoforms can be enhanced by insulin or cellular stress executed by viral transformation, topoisomerase inhibitor treatment, or γ-irradiation. Regulation of enzymatic activity is also possible on the protein level, *e.g.*, by modulation of subcellular localization, interaction with other proteins, or (auto-)phosphorylation in particular targeting the C-terminal regulatory domain but also the kinase domain[20,112].

Most CK1 isoforms are localized in the cytosol. Only the CK1αL variant possessing a second nuclear localization signal in the L-exon can be localized to the nucleus[113]. Due to C-terminal palmitoylation, CK1γ can be associated with the plasma membrane [114-116]. By modulation of subcellular localization, CK1 isoforms can be brought in proximity with different substrate pools. Substrate recognition motifs for CK1 can generally be found on most cellular proteins and, to date, more than 150 substrates being phosphorylated by CK1 isoforms at least *in vitro* have been reported[20]. Thereby, CK1 shows strong preferences for acidic or phospho-primed substrates presenting the canonical consensus sequence (phospho-Ser/phospho-Thr-X-X-(X)-Ser/Thr). In addition, several alternative noncanonical motifs targeted by CK1 have been described[117,118].

The broad range of substrates phosphorylated by CK1 gives a hint of the numerous cellular processes potentially regulated by CK1 family members. These processes involve cell proliferation and differentiation, DNA processing and repair, as well as

cytoskeleton maintenance just to name few of them. In particular, essential signal transduction pathways involving CK1-mediated regulation include Wnt and Hedgehog (Hh) signaling as well as regulation of circadian rhythm[20]. Consequently, deregulation or dysfunction of CK1 isoforms involved in regulation of these signaling pathways can result in deregulated signal transduction and subsequent development of pathological states.

Relevant CK1-related pathways in cancer

CK1 isoforms have been implicated in several signaling pathways such as the canonical and noncanonical Wnt as well as Hh and Hippo signaling pathways, which play an important role in tissue development, growth, and homeostasis[119-122]. Aberrant signaling as well as mutations of key regulator proteins of these pathways can lead to various cancer entities[123-127]. The connection between CK1 and cancer has been strengthened through the discovery of their targets such as β-catenin, p53, and mouse double minute homologue 2 and 4, which hold important roles as key regulators in signaling pathways and are generally thought to be involved in cancer development (Figure 1)[128,129]. Considering the reported CK1-mediated phosphorylation of numerous substrates essential for signal transduction, it is not surprising that CK1-specific mutations resulting in altered expression levels or enzyme activity are likely to be accompanied by dramatic changes affecting CK1-regulated signal transduction pathways.

As one of the best characterized CK1-regulated processes, the Wnt signaling pathway has an important regulatory role in cell proliferation, differentiation, and cell polarity[120,130-133]. Altered expression levels of key regulators within the pathway are associated to oncogenesis, both through increased expression of positive regulators and decreased expression of negative regulators[134-137]. Several studies showed that all CK1 isoforms are implicated in the Wnt signaling pathway and either exert positive or negative regulatory functions, respectively[14]. Acting as positive regulators of the canonical Wnt signaling pathway, CK1γ, δ, and ε were found to initiate the transcription of proto-oncogenes like cyclin D1 and c-myc resulting in increased cell proliferation and cell survival[13,138,139]. For instance, mutations within the C-terminal region of CK1δ were shown to alter its physiological role, increase the oncogenic potential, and promote colonic adenoma development[140]. Additionally, CK1 isoforms exhibit oncogenic characteristics associated to the inhibition of apoptotic processes. This assumption is supported by the findings that CK1δ and CK1ε contribute to the switching mechanism between the canonical and the non-canonical Wnt/Rac1/JNK pathway, where they may favor the canonical Wnt pathway to the detriment of JNK-mediated apoptosis[17,141]. In many Wnt-driven cancers, CK1α protein expression is suppressed, leading to an activation of proliferative processes *via* the Wnt pathway. In addition, the absence of CK1α leads to a critical involvement of p53 in controlling invasiveness, which was shown in a model for colon cancer[15].

The importance of CK1 isoforms within various signaling pathways is strengthened by reports linking CK1 to phosphorylation of components in Hh signaling pathway. Although the activity of the Hh signaling pathway is reduced in adulthood, it is critical for embryonic development, organogenesis, and maintenance of healthy adult cells[119]. In the adult organism, Hh signaling contributes to the regulation of epithelial maintenance and tissue regeneration; consequently mutations and dysregulation of components of this signaling pathway promote tumorigenesis and cancer development[142-145]. As seen in Wnt signaling, CK1 isoforms appear to have contrasting effects on Hh signaling. Acting as a negative regulator, CK1 promotes proteolysis of GLI TF and prevents target gene transcription[146-148]. In order to fulfill its positive function, CK1α and G-protein coupled receptor kinase 2 phosphorylate the positive Hh regulator Smoothened homologue precursor, thereby inducing its active conformation[149].

The major functions of the Hippo pathway have been defined to correct organ maturation through restriction of organ size by regulating cell proliferation and apoptosis[150]. As such, dysregulated Hippo signaling can trigger tumorigenesis and cancer. CK1 isoforms have been proposed to regulate Hippo signaling through phosphorylation of a phosphodegron signal in Yes-associated protein after receiving priming phosphorylation by large tumor suppressors 1 and 2. As a result, the phospho-degron signal mediates recruitment of βTrCP ubiquitin ligase causing Yes-associated protein degradation and inhibition of cell growth and differentiation[150]. Additionally, a more recent publication proposed an interaction between the Wnt and the Hippo pathways mediated through CK1ε. In this context, the Hippo upstream kinase MST1 inhibits the Wnt signaling pathway by directly binding CK1ε and thereby suppressing phosphorylation of Disheveled[151].

Implication for CK1 involvement in pancreatic cancer

In a recent study analyzing messenger RNA-based gene expression data of the International Cancer Genome Consortium Pancreatic Cancer Australia cohort, high expression levels of CK1δ detected in patients with pancreatic cancer were correlated with poor survival. Increased expression of CK1δ could be found in patients with metastatic pancreatic carcinoma, and CK1δ expression was furthermore strongly correlated with the tumor grade[152]. This observation is in line with previous studies reporting upregulation of CK1 isoforms in PDAC in general[153,154] and describing increased expression of CK1δ and CK1ε in a patient cohort with higher-graded PDAC [155]. Cell line-specific elevated expression levels of CK1δ and/or CK1ε were also detected in various tumor cell lines[152,155]. Independent of the detected protein levels, CK1δ- and CK1ε-specific kinase activities in extracts obtained from various pancreatic cancer cell lines (MiaPaCa-2, BxPC3, PancTu-1, and Colo357) significantly differed from each other by up to six orders of magnitude[156].

In general, due to the involvement of CK1 isoforms in various pathways related to tumorigenesis, altered expression and/or activity levels of CK1 isoforms can be associated with increased oncogenic potential. Using the breast cancer cell line MCF7, a regulatory function of CK1ε has been identified in the Akt pathway[157]. This is of particular interest because Akt is frequently upregulated in PDAC[158,159]. In detail, CK1ε is able to inhibit protein phosphatase 2B, consequently resulting in increased Akt phosphorylation levels and enhanced Akt kinase activity. Inhibition of CK1ε in MCF7 cells by the small molecule inhibitor IC261 has been demonstrated to reduce Akt phosphorylation as well as Akt-mediated phosphorylation of GSK3β[157]. Quite similar findings could be made using PDAC cells. Also in this case, phosphorylation of Akt was reduced in response to treatment with IC261[160]. However, these results were only based on observations made in preliminary experiments, and effects were obtained by using extremely high concentrations of the rather unspecific early-stage inhibitor IC261.

Apart from altered expression and/or activity levels, mutations in the coding sequence for CK1 isoforms can also be associated with increased oncogenic functions of the resulting CK1 mutant proteins. Several mutations in *CSNK1D*, the gene coding for human protein kinase CK1δ, identified in different types of cancer (*e.g.*, colorectal carcinoma, lung squamous cell carcinoma, bladder urothelial carcinoma, and pancreatic carcinoma) were analyzed for their enzyme kinetic parameters and their sensitivity towards the treatment with several CK1-specific small molecule inhibitors. Among the tested mutants, hyperactive (*e.g.*, R127L and R127Q) as well as nearly inactive (*e.g.*, E247K and L252P) variants could be characterized. Especially, the hyperactive CK1δ mutant R127Q showed enhanced sensitivity towards the treatment with various CK1-specific inhibitors. The two tested CK1δ mutants exclusively detected in PDAC (Q399* and H414Y) only showed slightly reduced kinase activity when compared to wild-type CK1δ[161]. The online analysis tool cBioPortal for Cancer Genomics lists even more mutations detected in PDAC and affecting CK1 isoforms, among them frameshift deletions as well as nonsense and missense mutations[162-165]. However, these mutations have so far not been investigated for their oncogenic potential.

Unfortunately, no detailed information on the role of CK1 isoforms in formation of metastasis from primary tumors located in the pancreas has been available so far. In general, the zinc-finger TF Snail is phosphorylated by CK1ε and GSK3β in a hierarchical manner. Snail can promote epithelial-mesenchymal transition by repressing expression of E-cadherin but is degraded by the proteasome upon phosphorylation by CK1ε and GSK3β. Pharmacological inhibition (using the inhibitor IC261) or RNA interference-mediated downregulation of CK1δ inhibits phosphorylation of Snail and promotes cell migration[166]. In addition, reduced proliferation and invasion could be linked with CK1ε-mediated inhibition of Wnt/β-catenin signaling and downregulation of Wnt3a, β-catenin, proliferating cell nuclear antigen, and matrix metalloproteinase 9 in colorectal cancer cells[167].

Inhibitors of CK1 and their role for the treatment of pancreatic cancer

Results obtained from numerous studies conducted within the last 10-15 years characterized the protein kinases of the CK1 family as well-established drug targets. While early-stage small molecule inhibitors (*e.g.*, IC261[168]) only demonstrated low target selectivity, several recent-stage CK1-specific inhibitors with enhanced selectivity and improved potency in the nanomolar range are available to date (Table 2)[20,112,169]. So far, none of these inhibitors advanced to the stage of clinical trials, and the use of these compounds was limited to biochemical and cell culture-based testing or animal

Table 2 Casein kinase 1-specific small molecule inhibitors tested for treatment effects on pancreatic carcinoma cells *in vitro* and *in vivo*

Inhibitor	IC_{50} CK1 (μmol/L)	Observed effects in cell culture and *in vivo* data	Ref.
IC261	1.000 ± 0.30 (CK1δ)	Reduced growth of ASPC-1, BxPC3, Capan-1, Colo357, MiaPaCa-2, Panc-1, Panc89, and PancTu-1 at 1.25 μmol/L concentration of IC261; Subcutaneous xenograft model using PancTu-2: reduced tumor size with IC261 or gemcitabine (no synergism with gemcitabine), downregulation of anti-apoptotic genes/upregulation of cell cycle- and cell death-associated regulators; Notable off target effects (affecting the cytoskeleton and ion channels)	[155, 168, 182-184, 202]
compound 11b	0.004 ± 0.001 (CK1δ); 0.025 ± 0.004 (CK1ε); 0.010 (p38α)	Cytotoxic effects observed on Colo357 (EC_{50} = 3.5 ± 0.3 μmol/L) and Panc89 (1.5 ± 0.4 μmol/L)	[174]
compound 3c	1.600 (CK1δ/ε)	In a panel of cell lines only effective against Panc-1 (EC_{50} = 9.3 ± 0.0 μmol/L); Cytotoxic effects observed on A549 (lung carcinoma) and Hek293 (normal cells) significantly higher EC_{50} values	[175]
compound 2	0.070 ± 0.01 (CK1δkd); 0.520 ± 0.05 (CK1ε)	Cytotoxic effects observed on BxPC3 (EC_{50} = 0.11 ± 0.01 μmol/L), Colo357 (0.13 ± 0.02 μmol/L), MiaPaCa (0.26 ± 0.02 μmol/L), PancTu-1 (0.70 ± 0.02 μmol/L), and Panc-1 (0.35 ± 0.08 μmol/L); Cell line-specific effects observed in screening against a panel of 82 tumor cell lines	[178]
IWP-4	1.020 ± 0.13 (CK1δ); 7.070 ± 2.01 (CK1ε)	Cytotoxic effects observed on A818-6 (EC_{50} = 0.93 ± 0.07 μmol/L), MiaPaCa-2 (0.23 ± 0.01 μmol/L), Panc-1 (0.23 ± 0.02 μmol/L), Panc89 (0.58 ± 0.12 μmol/L), and Capan (0.23 ± 0.01 μmol/L); Inhibition of Wnt signaling (Wnt3A overexpression, autocrine/paracrine) with IC_{50} = 0.71 ± 0.38 μmol/L; Inhibition of Wnt signaling (Wnt3A-conditioned medium, autocrine/paracrine) with EC_{50} = 1.47 ± 0.55 μmol/L	
SR-3029	0.044 (CK1δ); 0.260 (CK1ε)	Cytotoxic effects observed on Panc-1 (EC_{50} = 0.023 μmol/L), MiaPaCa2 (0.370 μmol/L), and BxPC3 (0.131 μmol/L); Mouse pharmacokinetic studies with promising results for animal model use of SR-3029; Orthotopic xenograft model using Panc-1, reduced tumor size using SR-3029 and/or gemcitabine (synergism with gemcitabine due to upregulation of dCK)	[152, 180]

CK1: Casein kinase 1; EC_{50}: Half maximal effective concentration; IC_{50}: Half maximal inhibitory concentration.

models.

Quite recently, we characterized optimized 4,5-diarylimidazoles as highly effective ATP-competitive inhibitors of CK1δ. Substituted isoxazoles were originally designed as inhibitors of p38α MAPK, but they share the same pharmacophore moiety that is necessary to inhibit CK1δ[93,170-172]. Substituting the isoxazole scaffold with an imidazole scaffold resulted in the generation of highly potent dual-specific inhibitors of p38α MAPK and CK1δ[173]. By further optimizing these imidazole-based compounds, CK1 isoform-specific inhibitors with IC_{50} values in the low nanomolar range like compounds 11b [$IC_{50(CK1\delta)}$ = 4 nmol/L], 12a (19 nmol/L), and 16b (8 nmol/L) could be developed, which represent the most potent CK1δ-specific inhibitors described so far. Because IC_{50} values determined for the highly related isoform CK1ε are increased by six to 12 orders of magnitude (with 25, 227, and 81 nmol/L for 11b, 12a, and 16b, respectively), these compounds can also be considered to be selective for CK1δ. Compound 11b even demonstrated superior selectivity towards CK1δ among a panel of more than 321 protein kinases. However, full selectivity with respect to side-effects on p38α MAPK could still not be achieved for this set of compounds, but IC_{50} values determined for p38α are three-fold higher compared to CK1δ. Finally, 11b demonstrated significant effects on pancreatic cancer cell lines, with half maximal effective concentration (EC_{50}) values in the low micromolar range [$EC_{50(Colo357)}$ = 3.5 μmol/L, $EC_{50(Panc89)}$ = 1.5 μmol/L)[174].

Apart from isoxazole- and imidazole-derived molecules, the quinazoline-based inhibitors (N-(1H-pyrazol-3-yl)quinazolin-4-amines) 3c and 3d have been shown to inhibit CK1δ and ε ($IC_{50(CK1\delta/\varepsilon)}$ = 1.6 and 1.4 μmol/L, respectively). In a panel of human cancer cell lines, compound 3c even demonstrated selective cytotoxicity against the PDAC cell line Panc-1, with an EC_{50} value of 9.3 μmol/L [for all others no EC_{50} value could be determined (> 100 μmol/L), except for A549 with 29.7 and HEK293 with 71.1 μmol/L]. Compound 3d also demonstrated effects on Panc-1 cells but only with an extremely high EC_{50} value of 69.4 μmol/L. However, the mechanism of selectivity of the tested quinazoline-based inhibitor remains to be determined[175].

Within the last decade, several benzimidazole-based inhibitors have demonstrated significant inhibition of CK1δ variants and superior isoform selectivity over CK1δ. The series of compounds described by Leban *et al*[176] originates from piperidinyl-thiazoles originally designed to inhibit nuclear factor-κB[176]. Following modification, these compounds also demonstrated significant inhibition of CK1 family members. Most significant inhibition of CK1δ kinase domain (CK1δkd) with superior isoform selectivity over CK1ε could be determined for compound 5 ($IC_{50(CK1\delta kd)}$ = 29 nmol/L, $IC_{50(CK1\varepsilon)}$ = 199 nmol/L). Compound 5 also induced apoptosis in various tumor cell lines

with cell line-specific effects and only moderate levels of apoptosis in Colo357 pancreatic cancer cells (tested at 4 μmol/L concentration)[177]. As reported by Richter and colleagues[178], the highly related but structurally slightly different compound 1 showed three-fold stronger inhibition of CK1δkd (IC_{50} = 10 nmol/L). By further improving the physicochemical properties of this difluoro-dioxolo-benzoimidazole derivative, inhibitor potency *in vitro* could be maintained for modified compound 2 ($IC_{50(CK1\delta kd)}$ = 0.07 μmol/L, $IC_{50(CK1\varepsilon)}$ = 0.52 μmol/L) while significantly increasing the effects observed on a panel of cancer cell lines. In comparison to compound 1, the effects on cell viability were significantly increased for cell lines treated with compound 2, among them the pancreatic cancer cell lines BxPC3, Colo357, MiaPaCa, PancTu-1, and Panc-1 (see Table 2 for EC_{50} data)[178].

Being structurally related to benzimidazole-based inhibitors, compounds derived from inhibitors of Wnt production (IWP) have recently been described as CK1-specific inhibitors. IWP-2 and IWP-4 as well as the further optimized compound 19 displayed rather potent inhibition of CK1δkd *in vitro* ($IC_{50(CK1\delta)}$ = 0.32, 1.02, and 0.09 μmol/L for IWP-2, IWP-4, and compound 19) and also demonstrated significant effects on the proliferation of pancreatic cancer cell lines as determined for IWP-4-treated A818-6 (EC_{50} = 0.93 μmol/L), MiaPaCa (0.23 μmol/L), Panc-1 (0.23 μmol/L), and Panc89 (0.58 μmol/L) cells[179].

As a benzimidazole-based inhibitor containing a purine scaffold compound SR-3029 has been described as highly potent and selective inhibitor of CK1δ ($IC_{50(CK1\delta)}$ = 44 nmol/L, $IC_{50(CK1\varepsilon)}$ = 260 nmol/L)[180]. SR-3029 shows improved cellular activity on the human melanoma cell line A375 (EC_{50} = 86 nmol/L) and the triple-negative breast cancer cell line MDA-MB-231[181]. These results suggested favorable cell penetration for SR-3029, and mouse pharmacokinetic properties indicated that SR-3029 actually was sufficient for use in xenograft studies[180].

Recently, SR-3029 has been tested for its effects on the proliferation of PDAC cell lines Panc-1, MiaPaCa-2, and BxPC3, thereby obtaining EC_{50} values in the submicromolar range (23, 370, and 131 nmol/L, respectively). Furthermore, synergistic effects have been detected for the treatment of MiaPaCa-2 and Panc-1 cells with a combination of SR-3029 and gemcitabine, the standard of care used in treatment of locally advanced and metastatic PDAC. Same effects could be observed after silencing of CK1δ by small interfering RNA. The mechanism of synergy could be explained by upregulation of deoxycytidine kinase subsequent to inhibition of CK1δ by SR-3029, resulting in enhanced metabolism and anti-proliferative effects of gemcitabine. Anti-proliferative effects of SR-3029 and synergy with gemcitabine could also be observed *in vivo* by using an orthotopic xenotransplantation mouse model. Tumors obtained from injection of Panc-1 cells into the pancreas were significantly smaller after treatment with SR-3029 or gemcitabine, and tumor size was even more reduced after combination therapy[152].

In a previous xenotransplantation study, the early-stage CK1-specific inhibitor IC261 had already demonstrated therapeutic potential. Tumor cell growth of a panel of established pancreatic cancer cell lines (ASPC-1, BxPC3, Capan-1, Colo357, MiaPaCa-2, Panc-1, Panc89, and PancTu-1) was significantly reduced by treatment with 1.25 μmol/L IC261 *in vitro*, and the size of tumors obtained after subcutaneous injection of PancTu-2 cells was significantly smaller after treatment with IC261. In the tumor tissue, downregulation of several anti-apoptotic genes (*e.g.*, Bcl-2 family members) and upregulation of cell cycle- and cell death-associated regulators (*e.g.*, p21, ataxia-telangiectasia mutated kinase, checkpoint kinase 1) could be observed following treatment with IC261 or gemcitabine[155]. However, and in contrast to the above mentioned recent study by Vena and colleagues[152], IC261 failed to sensitize gemcitabine-resistant PancTu-1 cells to treatment with gemcitabine, and no synergistic or additive action in combination with gemcitabine could be demonstrated for IC261. This failure can be due to the unspecific effects meanwhile described for IC261. Apart from its specific action on CK1 family members, IC261 is able to bind tubulin with an affinity similar to the spindle poison colchicine. IC261 can therefore be considered as a microtubule polymerization inhibitor by directly exerting its effects on microtubules independent of CK1 blockage[182,183]. Moreover, within the concentration range necessary to block CK1 kinase activity, IC261 is also able to block voltage-gated sodium channels, and consequently, well-characterized recent-stage CK1-specific inhibitor compounds like SR-3029 should be used for targeting CK1 isoforms instead of using the unspecific early-stage inhibitor IC261[184].

CONCLUSION

In recent years, there has been a lot of evidence for the involvement of stress-activated kinases like JNK and p38 but also CK1 in the pathogenesis of pancreatic cancer. Furthermore, remarkable progress has been made in designing specific small molecule inhibitors to effectively target these kinases *in vitro* and *in vivo* and to reduce off-target effects. Interestingly, due to similarities in protein structure, some inhibitor compounds even demonstrate dual inhibition of p38 and CK1 isoforms. However, further mechanisms and benefits from dual kinase inhibition have not been studied in detail. Furthermore, conclusive results from using specific inhibitors in clinical trials remain to be obtained, and knowledge on the interplay of these inhibitors with standard of care chemotherapeutics needs to be acquired in future studies.

REFERENCES

1 **Hidalgo M**, Cascinu S, Kleeff J, Labianca R, Löhr JM, Neoptolemos J, Real FX, Van Laethem JL, Heinemann V. Addressing the challenges of pancreatic cancer: future directions for improving outcomes. *Pancreatology* 2015; **15**: 8-18 [PMID: 25547205 DOI: 10.1016/j.pan.2014.10.001]

2 **Rahib L**, Smith BD, Aizenberg R, Rosenzweig AB, Fleshman JM, Matrisian LM. Projecting cancer incidence and deaths to 2030: the unexpected burden of thyroid, liver, and pancreas cancers in the United States. *Cancer Res* 2014; **74**: 2913-2921 [PMID: 24840647 DOI: 10.1158/0008-5472.CAN-14-0155]

3 **Hezel AF**, Kimmelman AC, Stanger BZ, Bardeesy N, Depinho RA. Genetics and biology of pancreatic ductal adenocarcinoma. *Genes Dev* 2006; **20**: 1218-1249 [PMID: 16702400 DOI: 10.1101/gad.1415606]

4 **Turjanski AG**, Vaqué JP, Gutkind JS. MAP kinases and the control of nuclear events. *Oncogene* 2007; **26**: 3240-3253 [PMID: 17496919 DOI: 10.1038/sj.onc.1210415]

5 **Zhang W**, Liu HT. MAPK signal pathways in the regulation of cell proliferation in mammalian cells. *Cell Res* 2002; **12**: 9-18 [PMID: 11942415 DOI: 10.1038/sj.cr.7290105]

6 **Pearson G**, Robinson F, Beers Gibson T, Xu BE, Karandikar M, Berman K, Cobb MH. Mitogen-activated protein (MAP) kinase pathways: regulation and physiological functions. *Endocr Rev* 2001; **22**: 153-183 [PMID: 11294822 DOI: 10.1210/edrv.22.2.0428]

7 **Avruch J**, Khokhlatchev A, Kyriakis JM, Luo Z, Tzivion G, Vavvas D, Zhang XF. Ras activation of the Raf kinase: tyrosine kinase recruitment of the MAP kinase cascade. *Recent Prog Horm Res* 2001; **56**: 127-155 [PMID: 11237210 DOI: 10.1210/rp.56.1.127]

8 **Collisson EA**, Trejo CL, Silva JM, Gu S, Korkola JE, Heiser LM, Charles RP, Rabinovich BA, Hann B, Dankort D, Spellman PT, Phillips WA, Gray JW, McMahon M. A central role for RAF→MEK→ERK signaling in the genesis of pancreatic ductal adenocarcinoma. *Cancer Discov* 2012; **2**: 685-693 [PMID: 22628411 DOI: 10.1158/2159-8290.CD-11-0347]

9 **Hayes TK**, Neel NF, Hu C, Gautam P, Chenard M, Long B, Aziz M, Kassner M, Bryant KL, Pierobon M, Marayati R, Kher S, George SD, Xu M, Wang-Gillam A, Samatar AA, Maitra A, Wennerberg K, Petricoin EF 3rd, Yin HH, Nelkin B, Cox AD, Yeh JJ, Der CJ. Long-Term ERK Inhibition in KRAS-Mutant Pancreatic Cancer Is Associated with MYC Degradation and Senescence-like Growth Suppression. *Cancer Cell* 2016; **29**: 75-89 [PMID: 26725216 DOI: 10.1016/j.ccell.2015.11.011]

10 **Dhillon AS**, Hagan S, Rath O, Kolch W. MAP kinase signalling pathways in cancer. *Oncogene* 2007; **26**: 3279-3290 [PMID: 17496922 DOI: 10.1038/sj.onc.1210421]

11 **He TC**, Sparks AB, Rago C, Hermeking H, Zawel L, da Costa LT, Morin PJ, Vogelstein B, Kinzler KW. Identification of c-MYC as a target of the APC pathway. *Science* 1998; **281**: 1509-1512 [PMID: 9727977 DOI: 10.1126/science.281.5382.1509]

12 **Tetsu O**, McCormick F. Beta-catenin regulates expression of cyclin D1 in colon carcinoma cells. *Nature* 1999; **398**: 422-426 [PMID: 10201372 DOI: 10.1038/18884]

13 **McKay RM**, Peters JM, Graff JM. The casein kinase I family in Wnt signaling. *Dev Biol* 2001; **235**: 388-396 [PMID: 11437445 DOI: 10.1006/dbio.2001.0308]

14 **Price MA**. CKI, there's more than one: casein kinase I family members in Wnt and Hedgehog signaling. *Genes Dev* 2006; **20**: 399-410 [PMID: 16481469 DOI: 10.1101/gad.1394306]

15 **Elyada E**, Pribluda A, Goldstein RE, Morgenstern Y, Brachya G, Cojocaru G, Snir-Alkalay I, Burstain I, Haffner-Krausz R, Jung S, Wiener Z, Alitalo K, Oren M, Pikarsky E, Ben-Neriah Y. CKI α ablation highlights a critical role for p53 in invasiveness control. *Nature* 2011; **470**: 409-413 [PMID: 21331045 DOI: 10.1038/nature09673]

16 **Zhang Y**, Qiu WJ, Liu DX, Neo SY, He X, Lin SC. Differential molecular assemblies underlie the dual function of Axin in modulating the WNT and JNK pathways. *J Biol Chem* 2001; **276**: 32152-32159 [PMID: 11408485 DOI: 10.1074/jbc.M104451200]

17 **Cong F**, Schweizer L, Varmus H. Casein kinase Iepsilon modulates the signaling specificities of dishevelled. *Mol Cell Biol* 2004; **24**: 2000-2011 [PMID: 14966280 DOI: 10.1128/mcb.24.5.2000-2011.2004]

18 **Desagher S**, Osen-Sand A, Montessuit S, Magnenat E, Vilbois F, Hochmann A, Journot L, Antonsson B, Martinou JC. Phosphorylation of bid by casein kinases I and II regulates its cleavage by caspase 8. *Mol Cell* 2001; **8**: 601-611 [PMID: 11583622 DOI: 10.1016/s1097-2765(01)00335-5]

19 **Knippschild U**, Milne DM, Campbell LE, DeMaggio AJ, Christenson E, Hoekstra MF, Meek DW. p53 is phosphorylated *in vitro* and *in vivo* by the delta and epsilon isoforms of casein kinase 1 and enhances the level of casein kinase 1 delta in response to topoisomerase-directed drugs. *Oncogene* 1997; **15**: 1727-1736 [PMID: 9349507 DOI: 10.1038/sj.onc.1201541]

20 **Xu P**, Ianes C, Gärtner F, Liu C, Burster T, Bakulev V, Rachidi N, Knippschild U, Bischof J. Structure, regulation, and (patho-)physiological functions of the stress-induced protein kinase CK1 delta (CSNK1D). *Gene* 2019; **715**: 144005 [PMID: 31376410 DOI: 10.1016/j.gene.2019.144005]

21 **Carrino M**, Quotti Tubi L, Fregnani A, Canovas Nunes S, Barilà G, Trentin L, Zambello R, Semenzato G, Manni S, Piazza F. Prosurvival autophagy is regulated by protein kinase CK1 alpha in multiple myeloma. *Cell Death Discov* 2019; **5**: 98 [PMID: 31123604 DOI: 10.1038/s41420-019-0179-1]

22 **Kyriakis JM**, Avruch J. pp54 microtubule-associated protein 2 kinase. A novel serine/threonine protein kinase regulated by phosphorylation and stimulated by poly-L-lysine. *J Biol Chem* 1990; **265**: 17355-17363 [PMID: 2170374]

23 **Davis RJ**. Signal transduction by the JNK group of MAP kinases. In: Letts LG, Morgan DW. Inflammatory Processes. Basel: Birkhäuser, 2000: 13-21

24 **Gupta S**, Barrett T, Whitmarsh AJ, Cavanagh J, Sluss HK, Dérijard B, Davis RJ. Selective interaction of JNK protein kinase isoforms with transcription factors. *EMBO J* 1996; **15**: 2760-2770 [PMID: 8654373]

25 **Ip YT**, Davis RJ. Signal transduction by the c-Jun N-terminal kinase (JNK)--from inflammation to development. *Curr Opin Cell Biol* 1998; **10**: 205-219 [PMID: 9561845 DOI: 10.1016/s0955-0674(98)80143-9]

26 **Dérijard B**, Hibi M, Wu IH, Barrett T, Su B, Deng T, Karin M, Davis RJ. JNK1: a protein kinase stimulated by UV light and Ha-Ras that binds and phosphorylates the c-Jun activation domain. *Cell* 1994; **76**: 1025-1037 [PMID: 8137421 DOI: 10.1016/0092-8674(94)90380-8]

27 **Tournier C**, Dong C, Turner TK, Jones SN, Flavell RA, Davis RJ. MKK7 is an essential component of the JNK signal transduction pathway activated by proinflammatory cytokines. *Genes Dev* 2001; **15**: 1419-1426 [PMID: 11390361 DOI: 10.1101/gad.888501]

28 **Tournier C**, Whitmarsh AJ, Cavanagh J, Barrett T, Davis RJ. The MKK7 gene encodes a group of c-Jun NH2-terminal kinase kinases. *Mol Cell Biol* 1999; **19**: 1569-1581 [PMID: 9891090 DOI: 10.1128/mcb.19.2.1569]

29 **Johnson GL**, Nakamura K. The c-jun kinase/stress-activated pathway: regulation, function and role in human disease. *Biochim Biophys Acta* 2007; **1773**: 1341-1348 [PMID: 17306896 DOI: 10.1016/j.bbamcr.2006.12.009]

30 **Zeke A**, Misheva M, Reményi A, Bogoyevitch MA. JNK Signaling: Regulation and Functions Based on Complex Protein-Protein Partnerships. *Microbiol Mol Biol Rev* 2016; **80**: 793-835 [PMID: 27466283 DOI: 10.1128/MMBR.00043-14]

31 **Morrison DK**, Davis RJ. Regulation of MAP kinase signaling modules by scaffold proteins in mammals. *Annu Rev Cell Dev Biol* 2003; **19**: 91-118 [PMID: 14570565 DOI: 10.1146/annurev.cellbio.19.111401.091942]

32 **Sheridan DL**, Kong Y, Parker SA, Dalby KN, Turk BE. Substrate discrimination among mitogen-activated protein kinases through distinct docking sequence motifs. *J Biol Chem* 2008; **283**: 19511-19520 [PMID: 18482985 DOI: 10.1074/jbc.M801074200]

33 **Coulthard LR**, White DE, Jones DL, McDermott MF, Burchill SA. p38(MAPK): stress responses from molecular mechanisms to therapeutics. *Trends Mol Med* 2009; **15**: 369-379 [PMID: 19665431 DOI: 10.1016/j.molmed.2009.06.005]

34 **Han J**, Lee JD, Bibbs L, Ulevitch RJ. A MAP kinase targeted by endotoxin and hyperosmolarity in mammalian cells. *Science* 1994; **265**: 808-811 [PMID: 7914033 DOI: 10.1126/science.7914033]

35 **Cuadrado A**, Nebreda AR. Mechanisms and functions of p38 MAPK signalling. *Biochem J* 2010; **429**: 403-417 [PMID: 20626350 DOI: 10.1042/BJ20100323]

36 **Raingeaud J**, Gupta S, Rogers JS, Dickens M, Han J, Ulevitch RJ, Davis RJ. Pro-inflammatory cytokines and environmental stress cause p38 mitogen-activated protein kinase activation by dual phosphorylation on tyrosine and threonine. *J Biol Chem* 1995; **270**: 7420-7426 [PMID: 7535770 DOI: 10.1074/jbc.270.13.7420]

37 **Alonso G**, Ambrosino C, Jones M, Nebreda AR. Differential activation of p38 mitogen-activated protein kinase isoforms depending on signal strength. *J Biol Chem* 2000; **275**: 40641-40648 [PMID: 11010976 DOI: 10.1074/jbc.M007835200]

38 **Enslen H**, Raingeaud J, Davis RJ. Selective activation of p38 mitogen-activated protein (MAP) kinase isoforms by the MAP kinase kinases MKK3 and MKK6. *J Biol Chem* 1998; **273**: 1741-1748 [PMID: 9430721 DOI: 10.1074/jbc.273.3.1741]

39 **Dérijard B**, Raingeaud J, Barrett T, Wu IH, Han J, Ulevitch RJ, Davis RJ. Independent human MAP-kinase signal transduction pathways defined by MEK and MKK isoforms. *Science* 1995; **267**: 682-685 [PMID: 7839144 DOI: 10.1126/science.7839144]

40 **Brancho D**, Tanaka N, Jaeschke A, Ventura JJ, Kelkar N, Tanaka Y, Kyuuma M, Takeshita T, Flavell RA, Davis RJ. Mechanism of p38 MAP kinase activation in vivo. *Genes Dev* 2003; **17**: 1969-1978 [PMID: 12893778 DOI: 10.1101/gad.1107303]

41 **Salvador JM**, Mittelstadt PR, Guszczynski T, Copeland TD, Yamaguchi H, Appella E, Fornace AJ Jr, Ashwell JD. Alternative p38 activation pathway mediated by T cell receptor-proximal tyrosine kinases. *Nat Immunol* 2005; **6**: 390-395 [PMID: 15735648 DOI: 10.1038/ni1177]

42 **Igea A**, Nebreda AR. The Stress Kinase p38α as a Target for Cancer Therapy. *Cancer Res* 2015; **75**: 3997-4002 [PMID: 26377941 DOI: 10.1158/0008-5472.CAN-15-0173]

43 **Tournier C**. The 2 Faces of JNK Signaling in Cancer. *Genes Cancer* 2013; **4**: 397-400 [PMID: 24349637 DOI: 10.1177/1947601913486349]

44 **Wagner EF**, Nebreda AR. Signal integration by JNK and p38 MAPK pathways in cancer development. *Nat Rev Cancer* 2009; **9**: 537-549 [PMID: 19629069 DOI: 10.1038/nrc2694]

45 **Martínez-Limón A**, Joaquin M, Caballero M, Posas F, de Nadal E. The p38 Pathway: From Biology to Cancer Therapy. *Int J Mol Sci* 2020; **21** [PMID: 32168915 DOI: 10.3390/ijms21061913]

46 **Sabapathy K**, Hochedlinger K, Nam SY, Bauer A, Karin M, Wagner EF. Distinct roles for JNK1 and JNK2 in regulating JNK activity and c-Jun-dependent cell proliferation. *Mol Cell* 2004; **15**: 713-725 [PMID: 15350216 DOI: 10.1016/j.molcel.2004.08.028]

47 **Fuchs SY**, Dolan L, Davis RJ, Ronai Z. Phosphorylation-dependent targeting of c-Jun ubiquitination by Jun N-kinase. *Oncogene* 1996; **13**: 1531-1535 [PMID: 8875991]

48 **Johnson R**, Spiegelman B, Hanahan D, Wisdom R. Cellular transformation and malignancy induced by ras require c-jun. *Mol Cell Biol* 1996; **16**: 4504-4511 [PMID: 8754851 DOI: 10.1128/mcb.16.8.4504]

49 **Smeal T**, Binetruy B, Mercola DA, Birrer M, Karin M. Oncogenic and transcriptional cooperation with Ha-Ras requires phosphorylation of c-Jun on serines 63 and 73. *Nature* 1991; **354**: 494-496 [PMID: 1749429 DOI: 10.1038/354494a0]

50 **Cellurale C**, Sabio G, Kennedy NJ, Das M, Barlow M, Sandy P, Jacks T, Davis RJ. Requirement of c-Jun NH(2)-terminal kinase for Ras-initiated tumor formation. *Mol Cell Biol* 2011; **31**: 1565-1576 [PMID: 21282468 DOI: 10.1128/MCB.01122-10]

51 **Nielsen C**, Thastrup J, Bøttzauw T, Jäättelä M, Kallunki T. c-Jun NH2-terminal kinase 2 is required for Ras transformation independently of activator protein 1. *Cancer Res* 2007; **67**: 178-185 [PMID: 17210697 DOI: 10.1158/0008-5472.CAN-06-2801]

52 **Kennedy NJ**, Sluss HK, Jones SN, Bar-Sagi D, Flavell RA, Davis RJ. Suppression of Ras-stimulated transformation by the JNK signal transduction pathway. *Genes Dev* 2003; **17**: 629-637 [PMID: 12629045 DOI: 10.1101/gad.1062903]

53 **Ventura JJ**, Hübner A, Zhang C, Flavell RA, Shokat KM, Davis RJ. Chemical genetic analysis of the time course of signal transduction by JNK. *Mol Cell* 2006; **21**: 701-710 [PMID: 16507367 DOI: 10.1016/j.molcel.2006.01.018]

54 **Hui L**, Zatloukal K, Scheuch H, Stepniak E, Wagner EF. Proliferation of human HCC cells and chemically induced mouse liver cancers requires JNK1-dependent p21 downregulation. *J Clin Invest* 2008; **118**: 3943-3953 [PMID: 19033664 DOI: 10.1172/JCI37156]

55 **Sakurai T**, Maeda S, Chang L, Karin M. Loss of hepatic NF-kappa B activity enhances chemical hepatocarcinogenesis through sustained c-Jun N-terminal kinase 1 activation. *Proc Natl Acad Sci USA* 2006; **103**: 10544-10551 [PMID: 16807293 DOI: 10.1073/pnas.0603499103]

56 **She QB**, Chen N, Bode AM, Flavell RA, Dong Z. Deficiency of c-Jun-NH(2)-terminal kinase-1 in mice enhances skin tumor development by 12-O-tetradecanoylphorbol-13-acetate. *Cancer Res* 2002; **62**: 1343-1348 [PMID: 11888903]

57 **Ambrosino C**, Nebreda AR. Cell cycle regulation by p38 MAP kinases. *Biol Cell* 2001; **93**: 47-51 [PMID: 11730321 DOI: 10.1016/s0248-4900(01)01124-8]

58 **Thoms HC**, Dunlop MG, Stark LA. p38-mediated inactivation of cyclin D1/cyclin-dependent kinase 4 stimulates nucleolar translocation of RelA and apoptosis in colorectal cancer cells. *Cancer Res* 2007; **67**: 1660-1669 [PMID: 17308107 DOI: 10.1158/0008-5472.CAN-06-1038]

59 **Bulavin DV**, Demidov ON, Saito S, Kauraniemi P, Phillips C, Amundson SA, Ambrosino C, Sauter G, Nebreda AR, Anderson CW, Kallioniemi A, Fornace AJ Jr, Appella E. Amplification of PPM1D in human tumors abrogates p53 tumor-suppressor activity. *Nat Genet* 2002; **31**: 210-215 [PMID: 12021785 DOI: 10.1038/ng894]

60 **She QB**, Bode AM, Ma WY, Chen NY, Dong Z. Resveratrol-induced activation of p53 and apoptosis is mediated by extracellular-signal-regulated protein kinases and p38 kinase. *Cancer Res* 2001; **61**: 1604-1610 [PMID: 11245472]

61 **Gubern A**, Joaquin M, Marquès M, Maseres P, Garcia-Garcia J, Amat R, González-Nuñez D, Oliva B, Real FX, de Nadal E, Posas F. The N-Terminal Phosphorylation of RB by p38 Bypasses Its Inactivation by CDKs and Prevents Proliferation in Cancer Cells. *Mol Cell* 2016; **64**: 25-36 [PMID: 27642049 DOI: 10.1016/j.molcel.2016.08.015]

62 **Philips A**, Roux P, Coulon V, Bellanger JM, Vié A, Vignais ML, Blanchard JM. Differential effect of Rac and Cdc42 on p38 kinase activity and cell cycle progression of nonadherent primary mouse fibroblasts. *J Biol Chem* 2000; **275**: 5911-5917 [PMID: 10681583 DOI: 10.1074/jbc.275.8.5911]

63 **Wang S**, Nath N, Minden A, Chellappan S. Regulation of Rb and E2F by signal transduction cascades: divergent effects of JNK1 and p38 kinases. *EMBO J* 1999; **18**: 1559-1570 [PMID: 10075927 DOI: 10.1093/emboj/18.6.1559]

64 **Thornton TM**, Rincon M. Non-classical p38 map kinase functions: cell cycle checkpoints and survival. *Int J Biol Sci* 2009; **5**: 44-51 [PMID: 19159010 DOI: 10.7150/ijbs.5.44]

65 **Lee ER**, Kim JY, Kang YJ, Ahn JY, Kim JH, Kim BW, Choi HY, Jeong MY, Cho SG. Interplay

between PI3K/Akt and MAPK signaling pathways in DNA-damaging drug-induced apoptosis. *Biochim Biophys Acta* 2006; **1763**: 958-968 [PMID: 16905201 DOI: 10.1016/j.bbamcr.2006.06.006]

66 **Hernández Losa J**, Parada Cobo C, Guinea Viniegra J, Sánchez-Arevalo Lobo VJ, Ramón y Cajal S, Sánchez-Prieto R. Role of the p38 MAPK pathway in cisplatin-based therapy. *Oncogene* 2003; **22**: 3998-4006 [PMID: 12821934 DOI: 10.1038/sj.onc.1206608]

67 **Handra-Luca A**, Lesty C, Hammel P, Sauvanet A, Rebours V, Martin A, Fagard R, Fléjou JF, Faivre S, Bédossa P, Ruszniewski P, Couvelard A. Biological and prognostic relevance of mitogen-activated protein kinases in pancreatic adenocarcinoma. *Pancreas* 2012; **41**: 416-421 [PMID: 22158075 DOI: 10.1097/MPA.0b013e318238379d]

68 **Zhong Y**, Naito Y, Cope L, Naranjo-Suarez S, Saunders T, Hong SM, Goggins MG, Herman JM, Wolfgang CL, Iacobuzio-Donahue CA. Functional p38 MAPK identified by biomarker profiling of pancreatic cancer restrains growth through JNK inhibition and correlates with improved survival. *Clin Cancer Res* 2014; **20**: 6200-6211 [PMID: 24963048 DOI: 10.1158/1078-0432.CCR-13-2823]

69 **Amsterdam A**, Shpigner L, Raanan C, Schreiber L, Melzer E, Seger R. Dynamic distribution of ERK, p38 and JNK during the development of pancreatic ductal adenocarcinoma. *Acta Histochem* 2014; **116**: 1434-1442 [PMID: 25440531 DOI: 10.1016/j.acthis.2014.09.007]

70 **Tian X**, Traub B, Xie X, Zhou S, Henne-Bruns D, Knippschild U, Kornmann M. Opposing Oncogenic Functions of p38 Mitogen-activated Protein Kinase Alpha and Beta in Human Pancreatic Cancer Cells. *Anticancer Res* 2020; **40**: 5545-5556 [PMID: 32988878 DOI: 10.21873/anticanres.14567]

71 **Dreissigacker U**, Mueller MS, Unger M, Siegert P, Genze F, Gierschik P, Giehl K. Oncogenic K-Ras down-regulates Rac1 and RhoA activity and enhances migration and invasion of pancreatic carcinoma cells through activation of p38. *Cell Signal* 2006; **18**: 1156-1168 [PMID: 16257181 DOI: 10.1016/j.cellsig.2005.09.004]

72 **Choi BH**, Philips MR, Chen Y, Lu L, Dai W. K-Ras Lys-42 is crucial for its signaling, cell migration, and invasion. *J Biol Chem* 2018; **293**: 17574-17581 [PMID: 30228186 DOI: 10.1074/jbc.RA118.003723]

73 **Takahashi R**, Hirata Y, Sakitani K, Nakata W, Kinoshita H, Hayakawa Y, Nakagawa H, Sakamoto K, Hikiba Y, Ijichi H, Moses HL, Maeda S, Koike K. Therapeutic effect of c-Jun N-terminal kinase inhibition on pancreatic cancer. *Cancer Sci* 2013; **104**: 337-344 [PMID: 23237571 DOI: 10.1111/cas.12080]

74 **Lu W**, Wei W, de Bock GH, Zhou H, Li Q, Shen X. The roles of Wnt5a, JNK and paxillin in the occurrence of metastasis of pancreatic adenocarcinoma. *Int J Clin Oncol* 2014; **19**: 1011-1019 [PMID: 24395444 DOI: 10.1007/s10147-013-0648-0]

75 **Bracci PM**, Zhou M, Young S, Wiemels J. Serum autoantibodies to pancreatic cancer antigens as biomarkers of pancreatic cancer in a San Francisco Bay Area case-control study. *Cancer* 2012; **118**: 5384-5394 [PMID: 22517435 DOI: 10.1002/cncr.27538]

76 **Tian X**, Traub B, Shi J, Huber N, Schreiner S, Chen G, Zhou S, Henne-Bruns D, Knippschild U, Kornmann M. c-Jun N-terminal kinase 2 suppresses pancreatic cancer growth and invasion and is opposed by c-Jun N-terminal kinase 1. *Cancer Gene Ther* 2021 [PMID: 33526844 DOI: 10.1038/s41417-020-00290-5]

77 **Hirano T**, Shino Y, Saito T, Komoda F, Okutomi Y, Takeda A, Ishihara T, Yamaguchi T, Saisho H, Shirasawa H. Dominant negative MEKK1 inhibits survival of pancreatic cancer cells. *Oncogene* 2002; **21**: 5923-5928 [PMID: 12185592 DOI: 10.1038/sj.onc.1205643]

78 **Teng DH**, Perry WL 3rd, Hogan JK, Baumgard M, Bell R, Berry S, Davis T, Frank D, Frye C, Hattier T, Hu R, Jammulapati S, Janecki T, Leavitt A, Mitchell JT, Pero R, Sexton D, Schroeder M, Su PH, Swedlund B, Kyriakis JM, Avruch J, Bartel P, Wong AK, Tavtigian SV. Human mitogen-activated protein kinase kinase 4 as a candidate tumor suppressor. *Cancer Res* 1997; **57**: 4177-4182 [PMID: 9331070]

79 **Xin W**, Yun KJ, Ricci F, Zahurak M, Qiu W, Su GH, Yeo CJ, Hruban RH, Kern SE, Iacobuzio-Donahue CA. MAP2K4/MKK4 expression in pancreatic cancer: genetic validation of immunohistochemistry and relationship to disease course. *Clin Cancer Res* 2004; **10**: 8516-8520 [PMID: 15623633 DOI: 10.1158/1078-0432.CCR-04-0885]

80 **Wang L**, Pan Y, Dai JL. Evidence of MKK4 pro-oncogenic activity in breast and pancreatic tumors. *Oncogene* 2004; **23**: 5978-5985 [PMID: 15184866 DOI: 10.1038/sj.onc.1207802]

81 **Sato T**, Shibata W, Hikiba Y, Kaneta Y, Suzuki N, Ihara S, Ishii Y, Sue S, Kameta E, Sugimori M, Yamada H, Kaneko H, Sasaki T, Ishii T, Tamura T, Kondo M, Maeda S. c-Jun N-terminal kinase in pancreatic tumor stroma augments tumor development in mice. *Cancer Sci* 2017; **108**: 2156-2165 [PMID: 28837246 DOI: 10.1111/cas.13382]

82 **Alam MS**, Gaida MM, Bergmann F, Lasitschka F, Giese T, Giese NA, Hackert T, Hinz U, Hussain SP, Kozlov SV, Ashwell JD. Selective inhibition of the p38 alternative activation pathway in infiltrating T cells inhibits pancreatic cancer progression. *Nat Med* 2015; **21**: 1337-1343 [PMID: 26479921 DOI: 10.1038/nm.3957]

83 **Siddiqui MA**, Reddy PA. Small molecule JNK (c-Jun N-terminal kinase) inhibitors. *J Med Chem* 2010; **53**: 3005-3012 [PMID: 20146479 DOI: 10.1021/jm9003279]

84 **Haller V**, Nahidino P, Forster M, Laufer SA. An updated patent review of p38 MAP kinase inhibitors (2014-2019). *Expert Opin Ther Pat* 2020; **30**: 453-466 [PMID: 32228113 DOI: 10.1080/13543776.2020.1749263]

85 **Wang W**, Ma C, Mao Z, Li M. JNK inhibition as a potential strategy in treating Parkinson's disease. *Drug News Perspect* 2004; **17**: 646-654 [PMID: 15696229 DOI: 10.1358/dnp.2004.17.10.873916]

86 L. Clinical candidates of small molecule p38 MAPK inhibitors for inflammatory diseases. *MAP Kinase* 2016; **4** [DOI: 10.4081/mk.2015.5508]

87 **Lee JK**, Kim NJ. Recent Advances in the Inhibition of p38 MAPK as a Potential Strategy for the Treatment of Alzheimer's Disease. *Molecules* 2017; **22** [PMID: 28767069 DOI: 10.3390/molecules22081287]

88 Celgene Corporation. Phase 1 Study to Determine the Optimal Biologic Dose of CC-401 in Subjects With High-Risk Myeloid Leukemia. [accessed 2021 Jan 22]. In: ClinicalTrials.gov [Internet]. Bethesda (MD): U.S. National Library of Medicine. Available from: https://www.clinicaltrials.gov/ct2/show/study/NCT00126893 ClinicalTrials.gov Identifier: NCT00126893

89 **Messoussi A**, Feneyrolles C, Bros A, Deroide A, Daydé-Cazals B, Chevé G, Van Hijfte N, Fauvel B, Bougrin K, Yasri A. Recent progress in the design, study, and development of c-Jun N-terminal kinase inhibitors as anticancer agents. *Chem Biol* 2014; **21**: 1433-1443 [PMID: 25442375 DOI: 10.1016/j.chembiol.2014.09.007]

90 **Ding XZ**, Adrian TE. MEK/ERK-mediated proliferation is negatively regulated by P38 map kinase in the human pancreatic cancer cell line, PANC-1. *Biochem Biophys Res Commun* 2001; **282**: 447-453 [PMID: 11401480 DOI: 10.1006/bbrc.2001.4595]

91 **Henklova P**, Vrzal R, Papouskova B, Bednar P, Jancova P, Anzenbacherova E, Ulrichova J, Maurel P, Pavek P, Dvorak Z. SB203580, a pharmacological inhibitor of p38 MAP kinase transduction pathway activates ERK and JNK MAP kinases in primary cultures of human hepatocytes. *Eur J Pharmacol* 2008; **593**: 16-23 [PMID: 18655782 DOI: 10.1016/j.ejphar.2008.07.007]

92 **Birkenkamp KU**, Tuyt LM, Lummen C, Wierenga AT, Kruijer W, Vellenga E. The p38 MAP kinase inhibitor SB203580 enhances nuclear factor-kappa B transcriptional activity by a non-specific effect upon the ERK pathway. *Br J Pharmacol* 2000; **131**: 99-107 [PMID: 10960075 DOI: 10.1038/sj.bjp.0703534]

93 **Shanware NP**, Williams LM, Bowler MJ, Tibbetts RS. Non-specific *in vivo* inhibition of CK1 by the pyridinyl imidazole p38 inhibitors SB 203580 and SB 202190. *BMB Rep* 2009; **42**: 142-147 [PMID: 19336000 DOI: 10.5483/bmbrep.2009.42.3.142]

94 **Hazzalin CA**, Cano E, Cuenda A, Barratt MJ, Cohen P, Mahadevan LC. p38/RK is essential for stress-induced nuclear responses: JNK/SAPKs and c-Jun/ATF-2 phosphorylation are insufficient. *Curr Biol* 1996; **6**: 1028-1031 [PMID: 8805335 DOI: 10.1016/s0960-9822(02)00649-8]

95 **Wada T**, Stepniak E, Hui L, Leibbrandt A, Katada T, Nishina H, Wagner EF, Penninger JM. Antagonistic control of cell fates by JNK and p38-MAPK signaling. *Cell Death Differ* 2008; **15**: 89-93 [PMID: 17762881 DOI: 10.1038/sj.cdd.4402222]

96 **Stepniak E**, Ricci R, Eferl R, Sumara G, Sumara I, Rath M, Hui L, Wagner EF. c-Jun/AP-1 controls liver regeneration by repressing p53/p21 and p38 MAPK activity. *Genes Dev* 2006; **20**: 2306-2314 [PMID: 16912279 DOI: 10.1101/gad.390506]

97 **Nemoto S**, Sheng Z, Lin A. Opposing effects of Jun kinase and p38 mitogen-activated protein kinases on cardiomyocyte hypertrophy. *Mol Cell Biol* 1998; **18**: 3518-3526 [PMID: 9584192 DOI: 10.1128/mcb.18.6.3518]

98 **Zhang B**, Meng M, Xiang S, Cao Z, Xu X, Zhao Z, Zhang T, Chen B, Yang P, Li Y, Zhou Q. Selective activation of tumor-suppressive MAPKP signaling pathway by triptonide effectively inhibits pancreatic cancer cell tumorigenicity and tumor growth. *Biochem Pharmacol* 2019; **166**: 70-81 [PMID: 31075266 DOI: 10.1016/j.bcp.2019.05.010]

99 **Yang L**, Sun X, Ye Y, Lu Y, Zuo J, Liu W, Elcock A, Zhu S. p38α Mitogen-Activated Protein Kinase Is a Druggable Target in Pancreatic Adenocarcinoma. *Front Oncol* 2019; **9**: 1294 [PMID: 31828036 DOI: 10.3389/fonc.2019.01294]

100 **Li C**, Heidt DG, Dalerba P, Burant CF, Zhang L, Adsay V, Wicha M, Clarke MF, Simeone DM. Identification of pancreatic cancer stem cells. *Cancer Res* 2007; **67**: 1030-1037 [PMID: 17283135 DOI: 10.1158/0008-5472.CAN-06-2030]

101 **Hermann PC**, Huber SL, Herrler T, Aicher A, Ellwart JW, Guba M, Bruns CJ, Heeschen C. Distinct populations of cancer stem cells determine tumor growth and metastatic activity in human pancreatic cancer. *Cell Stem Cell* 2007; **1**: 313-323 [PMID: 18371365 DOI: 10.1016/j.stem.2007.06.002]

102 **Okada M**, Shibuya K, Sato A, Seino S, Suzuki S, Seino M, Kitanaka C. Targeting the K-Ras--JNK axis eliminates cancer stem-like cells and prevents pancreatic tumor formation. *Oncotarget* 2014; **5**: 5100-5112 [PMID: 24947996 DOI: 10.18632/oncotarget.2087]

103 **Okada M**, Kuramoto K, Takeda H, Watarai H, Sakaki H, Seino S, Seino M, Suzuki S, Kitanaka C. The novel JNK inhibitor AS602801 inhibits cancer stem cells *in vitro* and in vivo. *Oncotarget* 2016; **7**: 27021-27032 [PMID: 27027242 DOI: 10.18632/oncotarget.8395]

104 **Konno T**, Ninomiya T, Kohno T, Kikuchi S, Sawada N, Kojima T. c-Jun N-terminal kinase inhibitor SP600125 enhances barrier function and elongation of human pancreatic cancer cell line HPAC in a Ca-switch model. *Histochem Cell Biol* 2015; **143**: 471-479 [PMID: 25511417 DOI: 10.1007/s00418-014-1300-4]

105 **Suzuki S**, Okada M, Shibuya K, Seino M, Sato A, Takeda H, Seino S, Yoshioka T, Kitanaka C. JNK suppression of chemotherapeutic agents-induced ROS confers chemoresistance on pancreatic cancer stem cells. *Oncotarget* 2015; **6**: 458-470 [PMID: 25473894 DOI: 10.18632/oncotarget.2693]

106 **Gottesman MM**, Fojo T, Bates SE. Multidrug resistance in cancer: role of ATP-dependent transporters. *Nat Rev Cancer* 2002; **2**: 48-58 [PMID: 11902585 DOI: 10.1038/nrc706]

107 **Zhou J**, Liu M, Aneja R, Chandra R, Lage H, Joshi HC. Reversal of P-glycoprotein-mediated multidrug resistance in cancer cells by the c-Jun NH2-terminal kinase. *Cancer Res* 2006; **66**: 445-452 [PMID: 16397260 DOI: 10.1158/0008-5472.CAN-05-1779]

108 **Li Y**, Köpper F, Dobbelstein M. Inhibition of MAPKAPK2/MK2 facilitates DNA replication upon cancer cell treatment with gemcitabine but not cisplatin. *Cancer Lett* 2018; **428**: 45-54 [PMID: 29704518 DOI: 10.1016/j.canlet.2018.04.030]

109 **BURNETT G**, KENNEDY EP. The enzymatic phosphorylation of proteins. *J Biol Chem* 1954; **211**: 969-980 [PMID: 13221602]

110 **Pinna LA**, Baggio B, Moret V, Siliprandi N. Isolation and properties of a protein kinase from rat liver microsomes. *Biochim Biophys Acta* 1969; **178**: 199-201 [PMID: 5773455 DOI: 10.1016/0005-2744(69)90152-1]

111 **Manning G**, Whyte DB, Martinez R, Hunter T, Sudarsanam S. The protein kinase complement of the human genome. *Science* 2002; **298**: 1912-1934 [PMID: 12471243 DOI: 10.1126/science.1075762]

112 **Knippschild U**, Krüger M, Richter J, Xu P, García-Reyes B, Peifer C, Halekotte J, Bakulev V, Bischof J. The CK1 Family: Contribution to Cellular Stress Response and Its Role in Carcinogenesis. *Front Oncol* 2014; **4**: 96 [PMID: 24904820 DOI: 10.3389/fonc.2014.00096]

113 **Fu Z**, Chakraborti T, Morse S, Bennett GS, Shaw G. Four casein kinase I isoforms are differentially partitioned between nucleus and cytoplasm. *Exp Cell Res* 2001; **269**: 275-286 [PMID: 11570820 DOI: 10.1006/excr.2001.5324]

114 **Davidson G**, Wu W, Shen J, Bilic J, Fenger U, Stannek P, Glinka A, Niehrs C. Casein kinase 1 gamma couples Wnt receptor activation to cytoplasmic signal transduction. *Nature* 2005; **438**: 867-872 [PMID: 16341016 DOI: 10.1038/nature04170]

115 **Robinson LC**, Menold MM, Garrett S, Culbertson MR. Casein kinase I-like protein kinases encoded by YCK1 and YCK2 are required for yeast morphogenesis. *Mol Cell Biol* 1993; **13**: 2870-2881 [PMID: 8474447 DOI: 10.1128/mcb.13.5.2870]

116 **Vancura A**, Sessler A, Leichus B, Kuret J. A prenylation motif is required for plasma membrane localization and biochemical function of casein kinase I in budding yeast. *J Biol Chem* 1994; **269**: 19271-19278 [PMID: 8034689]

117 **Marin O**, Bustos VH, Cesaro L, Meggio F, Pagano MA, Antonelli M, Allende CC, Pinna LA, Allende JE. A noncanonical sequence phosphorylated by casein kinase 1 in beta-catenin may play a role in casein kinase 1 targeting of important signaling proteins. *Proc Natl Acad Sci USA* 2003; **100**: 10193-10200 [PMID: 12925738 DOI: 10.1073/pnas.1733909100]

118 **Kawakami F**, Suzuki K, Ohtsuki K. A novel consensus phosphorylation motif in sulfatide- and cholesterol-3-sulfate-binding protein substrates for CK1 in vitro. *Biol Pharm Bull* 2008; **31**: 193-200 [PMID: 18239272 DOI: 10.1248/bpb.31.193]

119 **Ingham PW**, McMahon AP. Hedgehog signaling in animal development: paradigms and principles. *Genes Dev* 2001; **15**: 3059-3087 [PMID: 11731473 DOI: 10.1101/gad.938601]

120 **Logan CY**, Nusse R. The Wnt signaling pathway in development and disease. *Annu Rev Cell Dev Biol* 2004; **20**: 781-810 [PMID: 15473860 DOI: 10.1146/annurev.cellbio.20.010403.113126]

121 **Pan D**. Hippo signaling in organ size control. *Genes Dev* 2007; **21**: 886-897 [PMID: 17437995 DOI: 10.1101/gad.1536007]

122 **Zhao B**, Tumaneng K, Guan KL. The Hippo pathway in organ size control, tissue regeneration and stem cell self-renewal. *Nat Cell Biol* 2011; **13**: 877-883 [PMID: 21808241 DOI: 10.1038/ncb2303]

123 **Bao Y**, Hata Y, Ikeda M, Withanage K. Mammalian Hippo pathway: from development to cancer and beyond. *J Biochem* 2011; **149**: 361-379 [PMID: 21324984 DOI: 10.1093/jb/mvr021]

124 **Clevers H**. Wnt/beta-catenin signaling in development and disease. *Cell* 2006; **127**: 469-480 [PMID: 17081971 DOI: 10.1016/j.cell.2006.10.018]

125 **Klaus A**, Birchmeier W. Wnt signalling and its impact on development and cancer. *Nat Rev Cancer* 2008; **8**: 387-398 [PMID: 18432252 DOI: 10.1038/nrc2389]

126 **Pan D**. The hippo signaling pathway in development and cancer. *Dev Cell* 2010; **19**: 491-505 [PMID: 20951342 DOI: 10.1016/j.devcel.2010.09.011]

127 **Rubin LL**, de Sauvage FJ. Targeting the Hedgehog pathway in cancer. *Nat Rev Drug Discov* 2006; **5**: 1026-1033 [PMID: 17139287 DOI: 10.1038/nrd2086]

128 **Knippschild U**, Gocht A, Wolff S, Huber N, Löhler J, Stöter M. The casein kinase 1 family: participation in multiple cellular processes in eukaryotes. *Cell Signal* 2005; **17**: 675-689 [PMID: 15722192 DOI: 10.1016/j.cellsig.2004.12.011]

129 **Knippschild U**, Wolff S, Giamas G, Brockschmidt C, Wittau M, Würl PU, Eismann T, Stöter M. The role of the casein kinase 1 (CK1) family in different signaling pathways linked to cancer development. *Onkologie* 2005; **28**: 508-514 [PMID: 16186692 DOI: 10.1159/000087137]

130 **Cadigan KM**, Nusse R. Wnt signaling: a common theme in animal development. *Genes Dev* 1997; **11**: 3286-3305 [PMID: 9407023 DOI: 10.1101/gad.11.24.3286]

131 **McMahon AP**, Moon RT. Ectopic expression of the proto-oncogene int-1 in Xenopus embryos leads to duplication of the embryonic axis. *Cell* 1989; **58**: 1075-1084 [PMID: 2673541 DOI: 10.1016/0092-8674(89)90506-0]

132 **Moon RT**, Brown JD, Torres M. WNTs modulate cell fate and behavior during vertebrate

development. *Trends Genet* 1997; **13**: 157-162 [PMID: 9097727 DOI: 10.1016/s0168-9525(97)01093-7]

133 **Rocheleau CE**, Downs WD, Lin R, Wittmann C, Bei Y, Cha YH, Ali M, Priess JR, Mello CC. Wnt signaling and an APC-related gene specify endoderm in early C. elegans embryos. *Cell* 1997; **90**: 707-716 [PMID: 9288750 DOI: 10.1016/s0092-8674(00)80531-0]

134 **Sinnberg T**, Menzel M, Kaesler S, Biedermann T, Sauer B, Nahnsen S, Schwarz M, Garbe C, Schittek B. Suppression of casein kinase 1alpha in melanoma cells induces a switch in beta-catenin signaling to promote metastasis. *Cancer Res* 2010; **70**: 6999-7009 [PMID: 20699366 DOI: 10.1158/0008-5472.CAN-10-0645]

135 **Xiang XJ**, Liu YW, Chen DD, Yu S. Differential expression of Dickkopf-1 among non-small cell lung cancer cells. *Mol Med Rep* 2015; **12**: 1935-1940 [PMID: 25901391 DOI: 10.3892/mmr.2015.3654]

136 **Powell SM**, Zilz N, Beazer-Barclay Y, Bryan TM, Hamilton SR, Thibodeau SN, Vogelstein B, Kinzler KW. APC mutations occur early during colorectal tumorigenesis. *Nature* 1992; **359**: 235-237 [PMID: 1528264 DOI: 10.1038/359235a0]

137 **Nagahata T**, Shimada T, Harada A, Nagai H, Onda M, Yokoyama S, Shiba T, Jin E, Kawanami O, Emi M. Amplification, up-regulation and over-expression of DVL-1, the human counterpart of the Drosophila disheveled gene, in primary breast cancers. *Cancer Sci* 2003; **94**: 515-518 [PMID: 12824876 DOI: 10.1111/j.1349-7006.2003.tb01475.x]

138 **Wong C**, Chen C, Wu Q, Liu Y, Zheng P. A critical role for the regulated wnt-myc pathway in naive T cell survival. *J Immunol* 2015; **194**: 158-167 [PMID: 25429066 DOI: 10.4049/jimmunol.1401238]

139 **Fusté NP**, Fernández-Hernández R, Cemeli T, Mirantes C, Pedraza N, Rafel M, Torres-Rosell J, Colomina N, Ferrezuelo F, Dolcet X, Garí E. Cytoplasmic cyclin D1 regulates cell invasion and metastasis through the phosphorylation of paxillin. *Nat Commun* 2016; **7**: 11581 [PMID: 27181366 DOI: 10.1038/ncomms11581]

140 **Tsai IC**, Woolf M, Neklason DW, Branford WW, Yost HJ, Burt RW, Virshup DM. Disease-associated casein kinase I delta mutation may promote adenomatous polyps formation *via* a Wnt/beta-catenin independent mechanism. *Int J Cancer* 2007; **120**: 1005-1012 [PMID: 17131344 DOI: 10.1002/ijc.22368]

141 **Zhang Y**, Qiu WJ, Chan SC, Han J, He X, Lin SC. Casein kinase I and casein kinase II differentially regulate axin function in Wnt and JNK pathways. *J Biol Chem* 2002; **277**: 17706-17712 [PMID: 11884395 DOI: 10.1074/jbc.M111982200]

142 **Beachy PA**, Karhadkar SS, Berman DM. Tissue repair and stem cell renewal in carcinogenesis. *Nature* 2004; **432**: 324-331 [PMID: 15549094 DOI: 10.1038/nature03100]

143 **Lindemann RK**. Stroma-initiated hedgehog signaling takes center stage in B-cell lymphoma. *Cancer Res* 2008; **68**: 961-964 [PMID: 18281468 DOI: 10.1158/0008-5472.CAN-07-5500]

144 **Ruiz i Altaba A**, Mas C, Stecca B. The Gli code: an information nexus regulating cell fate, stemness and cancer. *Trends Cell Biol* 2007; **17**: 438-447 [PMID: 17845852 DOI: 10.1016/j.tcb.2007.06.007]

145 **Wicking C**, Smyth I, Bale A. The hedgehog signalling pathway in tumorigenesis and development. *Oncogene* 1999; **18**: 7844-7851 [PMID: 10630637 DOI: 10.1038/sj.onc.1203282]

146 **Denef N**, Neubüser D, Perez L, Cohen SM. Hedgehog induces opposite changes in turnover and subcellular localization of patched and smoothened. *Cell* 2000; **102**: 521-531 [PMID: 10966113 DOI: 10.1016/s0092-8674(00)00056-8]

147 **Taipale J**, Cooper MK, Maiti T, Beachy PA. Patched acts catalytically to suppress the activity of Smoothened. *Nature* 2002; **418**: 892-897 [PMID: 12192414 DOI: 10.1038/nature00989]

148 **Varjosalo M**, Taipale J. Hedgehog: functions and mechanisms. *Genes Dev* 2008; **22**: 2454-2472 [PMID: 18794343 DOI: 10.1101/gad.1693608]

149 **Chen Y**, Sasai N, Ma G, Yue T, Jia J, Briscoe J, Jiang J. Sonic Hedgehog dependent phosphorylation by CK1α and GRK2 is required for ciliary accumulation and activation of smoothened. *PLoS Biol* 2011; **9**: e1001083 [PMID: 21695114 DOI: 10.1371/journal.pbio.1001083]

150 **Zhao B**, Li L, Tumaneng K, Wang CY, Guan KL. A coordinated phosphorylation by Lats and CK1 regulates YAP stability through SCF(beta-TRCP). *Genes Dev* 2010; **24**: 72-85 [PMID: 20048001 DOI: 10.1101/gad.1843810]

151 **Xu F**, Wang YL, Chang JJ, Du SC, Diao L, Jiang N, Wang HJ, Ma D, Zhang J. Mammalian sterile 20-like kinase 1/2 inhibits the Wnt/β-catenin signalling pathway by directly binding casein kinase 1ε. *Biochem J* 2014; **458**: 159-169 [PMID: 24180524 DOI: 10.1042/BJ20130986]

152 **Vena F**, Bayle S, Nieto A, Quereda V, Aceti M, Frydman SM, Sansil SS, Grant W, Monastyrskyi A, McDonald P, Roush WR, Teng M, Duckett D. Targeting Casein Kinase 1 Delta Sensitizes Pancreatic and Bladder Cancer Cells to Gemcitabine Treatment by Upregulating Deoxycytidine Kinase. *Mol Cancer Ther* 2020; **19**: 1623-1635 [PMID: 32430484 DOI: 10.1158/1535-7163.MCT-19-0997]

153 **Grützmann R**, Boriss H, Ammerpohl O, Lüttges J, Kalthoff H, Schackert HK, Klöppel G, Saeger HD, Pilarsky C. Meta-analysis of microarray data on pancreatic cancer defines a set of commonly dysregulated genes. *Oncogene* 2005; **24**: 5079-5088 [PMID: 15897887 DOI: 10.1038/sj.onc.1208696]

154 **Grützmann R**, Pilarsky C, Staub E, Schmitt AO, Foerder M, Specht T, Hinzmann B, Dahl E, Alldinger I, Rosenthal A, Ockert D, Saeger HD. Systematic isolation of genes differentially expressed in normal and cancerous tissue of the pancreas. *Pancreatology* 2003; **3**: 169-178 [PMID:

12748427 DOI: 10.1159/000070087]

155 **Brockschmidt C**, Hirner H, Huber N, Eismann T, Hillenbrand A, Giamas G, Radunsky B, Ammerpohl O, Bohm B, Henne-Bruns D, Kalthoff H, Leithäuser F, Trauzold A, Knippschild U. Anti-apoptotic and growth-stimulatory functions of CK1 delta and epsilon in ductal adenocarcinoma of the pancreas are inhibited by IC261 *in vitro* and in vivo. *Gut* 2008; **57**: 799-806 [PMID: 18203806 DOI: 10.1136/gut.2007.123695]

156 **Giamas G**, Hirner H, Shoshiashvili L, Grothey A, Gessert S, Kühl M, Henne-Bruns D, Vorgias CE, Knippschild U. Phosphorylation of CK1delta: identification of Ser370 as the major phosphorylation site targeted by PKA *in vitro* and in vivo. *Biochem J* 2007; **406**: 389-398 [PMID: 17594292 DOI: 10.1042/BJ20070091]

157 **Modak C**, Bryant P. Casein Kinase I epsilon positively regulates the Akt pathway in breast cancer cell lines. *Biochem Biophys Res Commun* 2008; **368**: 801-807 [PMID: 18262492 DOI: 10.1016/j.bbrc.2008.02.001]

158 **Bondar VM**, Sweeney-Gotsch B, Andreeff M, Mills GB, McConkey DJ. Inhibition of the phosphatidylinositol 3'-kinase-AKT pathway induces apoptosis in pancreatic carcinoma cells *in vitro* and in vivo. *Mol Cancer Ther* 2002; **1**: 989-997 [PMID: 12481421]

159 **Yamamoto S**, Tomita Y, Hoshida Y, Morooka T, Nagano H, Dono K, Umeshita K, Sakon M, Ishikawa O, Ohigashi H, Nakamori S, Monden M, Aozasa K. Prognostic significance of activated Akt expression in pancreatic ductal adenocarcinoma. *Clin Cancer Res* 2004; **10**: 2846-2850 [PMID: 15102693 DOI: 10.1158/1078-0432.ccr-02-1441]

160 **Modak C**, Chai J. Potential of casein kinase I in digestive cancer screening. *World J Gastrointest Oncol* 2009; **1**: 26-33 [PMID: 21160770 DOI: 10.4251/wjgo.v1.i1.26]

161 **Liu C**, Witt L, Ianes C, Bischof J, Bammert MT, Baier J, Kirschner S, Henne-Bruns D, Xu P, Kornmann M, Peifer C, Knippschild U. Newly Developed CK1-Specific Inhibitors Show Specifically Stronger Effects on CK1 Mutants and Colon Cancer Cell Lines. *Int J Mol Sci* 2019; **20** [PMID: 31817920 DOI: 10.3390/ijms20246184]

162 **Broad Institute TCGA Genome Data Analysis Center**. Analysis Overview for Pancreatic Adenocarcinoma (Primary solid tumor cohort) - 28 January 2016; 2016 [cited 2021 Jan 22]. Database: cBioPortal [Internet]. Available from: https://www.cbioportal.org/study/summary?id=paad_tcga

163 **Bailey P**, Chang DK, Nones K, Johns AL, Patch AM, Gingras MC, Miller DK, Christ AN, Bruxner TJ, Quinn MC, Nourse C, Murtaugh LC, Harliwong I, Idrisoglu S, Manning S, Nourbakhsh E, Wani S, Fink L, Holmes O, Chin V, Anderson MJ, Kazakoff S, Leonard C, Newell F, Waddell N, Wood S, Xu Q, Wilson PJ, Cloonan N, Kassahn KS, Taylor D, Quek K, Robertson A, Pantano L, Mincarelli L, Sanchez LN, Evers L, Wu J, Pinese M, Cowley MJ, Jones MD, Colvin EK, Nagrial AM, Humphrey ES, Chantrill LA, Mawson A, Humphris J, Chou A, Pajic M, Scarlett CJ, Pinho AV, Giry-Laterriere M, Rooman I, Samra JS, Kench JG, Lovell JA, Merrett ND, Toon CW, Epari K, Nguyen NQ, Barbour A, Zeps N, Moran-Jones K, Jamieson NB, Graham JS, Duthie F, Oien K, Hair J, Grützmann R, Maitra A, Iacobuzio-Donahue CA, Wolfgang CL, Morgan RA, Lawlor RT, Corbo V, Bassi C, Rusev B, Capelli P, Salvia R, Tortora G, Mukhopadhyay D, Petersen GM; Australian Pancreatic Cancer Genome Initiative, Munzy DM, Fisher WE, Karim SA, Eshleman JR, Hruban RH, Pilarsky C, Morton JP, Sansom OJ, Scarpa A, Musgrove EA, Bailey UM, Hofmann O, Sutherland RL, Wheeler DA, Gill AJ, Gibbs RA, Pearson JV, Biankin AV, Grimmond SM. Genomic analyses identify molecular subtypes of pancreatic cancer. *Nature* 2016; **531**: 47-52 [PMID: 26909576 DOI: 10.1038/nature16965]

164 **Cerami E**, Gao J, Dogrusoz U, Gross BE, Sumer SO, Aksoy BA, Jacobsen A, Byrne CJ, Heuer ML, Larsson E, Antipin Y, Reva B, Goldberg AP, Sander C, Schultz N. The cBio cancer genomics portal: an open platform for exploring multidimensional cancer genomics data. *Cancer Discov* 2012; **2**: 401-404 [PMID: 22588877 DOI: 10.1158/2159-8290.CD-12-0095]

165 **Gao J**, Aksoy BA, Dogrusoz U, Dresdner G, Gross B, Sumer SO, Sun Y, Jacobsen A, Sinha R, Larsson E, Cerami E, Sander C, Schultz N. Integrative analysis of complex cancer genomics and clinical profiles using the cBioPortal. *Sci Signal* 2013; **6**: pl1 [PMID: 23550210 DOI: 10.1126/scisignal.2004088]

166 **Xu Y**, Lee SH, Kim HS, Kim NH, Piao S, Park SH, Jung YS, Yook JI, Park BJ, Ha NC. Role of CK1 in GSK3beta-mediated phosphorylation and degradation of snail. *Oncogene* 2010; **29**: 3124-3133 [PMID: 20305697 DOI: 10.1038/onc.2010.77]

167 **Ye LC**, Jiang C, Bai J, Jiang J, Hong HF, Qiu LS. Knockdown of casein kinase 1e inhibits cell proliferation and invasion of colorectal cancer cells *via* inhibition of the wnt/β-catenin signaling. *J Biol Regul Homeost Agents* 2015; **29**: 307-315 [PMID: 26122218]

168 **Mashhoon N**, DeMaggio AJ, Tereshko V, Bergmeier SC, Egli M, Hoekstra MF, Kuret J. Crystal structure of a conformation-selective casein kinase-1 inhibitor. *J Biol Chem* 2000; **275**: 20052-20060 [PMID: 10749871 DOI: 10.1074/jbc.M001713200]

169 **Cozza G**, Pinna LA. Casein kinases as potential therapeutic targets. *Expert Opin Ther Targets* 2016; **20**: 319-340 [PMID: 26565594 DOI: 10.1517/14728222.2016.1091883]

170 **Peifer C**, Kinkel K, Abadleh M, Schollmeyer D, Laufer S. From five- to six-membered rings: 3,4-diarylquinolinone as lead for novel p38MAP kinase inhibitors. *J Med Chem* 2007; **50**: 1213-1221 [PMID: 17323937 DOI: 10.1021/jm061097o]

171 **Peifer C**, Urich R, Schattel V, Abadleh M, Röttig M, Kohlbacher O, Laufer S. Implications for selectivity of 3,4-diarylquinolinones as p38alphaMAP kinase inhibitors. *Bioorg Med Chem Lett*

2008; **18**: 1431-1435 [PMID: 18207396 DOI: 10.1016/j.bmcl.2007.12.073]

172 **Godl K**, Wissing J, Kurtenbach A, Habenberger P, Blencke S, Gutbrod H, Salassidis K, Stein-Gerlach M, Missio A, Cotten M, Daub H. An efficient proteomics method to identify the cellular targets of protein kinase inhibitors. *Proc Natl Acad Sci USA* 2003; **100**: 15434-15439 [PMID: 14668439 DOI: 10.1073/pnas.2535024100]

173 **Peifer C**, Abadleh M, Bischof J, Hauser D, Schattel V, Hirner H, Knippschild U, Laufer S. 3,4-Diaryl-isoxazoles and -imidazoles as potent dual inhibitors of p38alpha mitogen activated protein kinase and casein kinase 1delta. *J Med Chem* 2009; **52**: 7618-7630 [PMID: 19591487 DOI: 10.1021/jm9005127]

174 **Halekotte J**, Witt L, Ianes C, Krüger M, Bührmann M, Rauh D, Pichlo C, Brunstein E, Luxenburger A, Baumann U, Knippschild U, Bischof J, Peifer C. Optimized 4,5-Diarylimidazoles as Potent/Selective Inhibitors of Protein Kinase CK1δ and Their Structural Relation to p38α MAPK. *Molecules* 2017; **22** [PMID: 28338621 DOI: 10.3390/molecules22040522]

175 **Karthikeyan C**, Jharia P, Waiker DK, Nusbaum AC, Amawi H, Kirwen EM, Christman R, Arudra SKC, Meijer L, Tiwari AK, Trivedi P. N-(1H-Pyrazol-3-yl)quinazolin-4-amines as a novel class of casein kinase 1δ/ε inhibitors: Synthesis, biological evaluation and molecular modeling studies. *Bioorg Med Chem Lett* 2017; **27**: 2663-2667 [PMID: 28487075 DOI: 10.1016/j.bmcl.2017.04.080]

176 **Leban J**, Baierl M, Mies J, Trentinaglia V, Rath S, Kronthaler K, Wolf K, Gotschlich A, Seifert MH. A novel class of potent NF-kappaB signaling inhibitors. *Bioorg Med Chem Lett* 2007; **17**: 5858-5862 [PMID: 17869512 DOI: 10.1016/j.bmcl.2007.08.022]

177 **Bischof J**, Leban J, Zaja M, Grothey A, Radunsky B, Othersen O, Strobl S, Vitt D, Knippschild U. 2-Benzamido-N-(1H-benzo[d]imidazol-2-yl)thiazole-4-carboxamide derivatives as potent inhibitors of CK1δ/ε. *Amino Acids* 2012; **43**: 1577-1591 [PMID: 22331384 DOI: 10.1007/s00726-012-1234-x]

178 **Richter J**, Bischof J, Zaja M, Kohlhof H, Othersen O, Vitt D, Alscher V, Pospiech I, García-Reyes B, Berg S, Leban J, Knippschild U. Difluoro-dioxolo-benzoimidazol-benzamides as potent inhibitors of CK1δ and ε with nanomolar inhibitory activity on cancer cell proliferation. *J Med Chem* 2014; **57**: 7933-7946 [PMID: 25191940 DOI: 10.1021/jm500600b]

179 **García-Reyes B**, Witt L, Jansen B, Karasu E, Gehring T, Leban J, Henne-Bruns D, Pichlo C, Brunstein E, Baumann U, Wesseler F, Rathmer B, Schade D, Peifer C, Knippschild U. Discovery of Inhibitor of Wnt Production 2 (IWP-2) and Related Compounds As Selective ATP-Competitive Inhibitors of Casein Kinase 1 (CK1) δ/ε. *J Med Chem* 2018; **61**: 4087-4102 [PMID: 29630366 DOI: 10.1021/acs.jmedchem.8b00095]

180 **Bibian M**, Rahaim RJ, Choi JY, Noguchi Y, Schürer S, Chen W, Nakanishi S, Licht K, Rosenberg LH, Li L, Feng Y, Cameron MD, Duckett DR, Cleveland JL, Roush WR. Development of highly selective casein kinase 1δ/1ε (CK1δ/ε) inhibitors with potent antiproliferative properties. *Bioorg Med Chem Lett* 2013; **23**: 4374-4380 [PMID: 23787102 DOI: 10.1016/j.bmcl.2013.05.075]

181 **Rosenberg LH**, Lafitte M, Quereda V, Grant W, Chen W, Bibian M, Noguchi Y, Fallahi M, Yang C, Chang JC, Roush WR, Cleveland JL, Duckett DR. Therapeutic targeting of casein kinase 1δ in breast cancer. *Sci Transl Med* 2015; **7**: 318ra202 [PMID: 26676609 DOI: 10.1126/scitranslmed.aac8773]

182 **Cheong JK**, Nguyen TH, Wang H, Tan P, Voorhoeve PM, Lee SH, Virshup DM. IC261 induces cell cycle arrest and apoptosis of human cancer cells *via* CK1δ/ε and Wnt/β-catenin independent inhibition of mitotic spindle formation. *Oncogene* 2011; **30**: 2558-2569 [PMID: 21258417 DOI: 10.1038/onc.2010.627]

183 **Stöter M**, Krüger M, Banting G, Henne-Bruns D, Knippschild U. Microtubules depolymerization caused by the CK1 inhibitor IC261 may be not mediated by CK1 blockage. *PLoS One* 2014; **9**: e100090 [PMID: 24937750 DOI: 10.1371/journal.pone.0100090]

184 **Föhr KJ**, Knippschild U, Herkommer A, Fauler M, Peifer C, Georgieff M, Adolph O. State-dependent block of voltage-gated sodium channels by the casein-kinase 1 inhibitor IC261. *Invest New Drugs* 2017; **35**: 277-289 [PMID: 28164251 DOI: 10.1007/s10637-017-0429-0]

185 **Bennett BL**, Sasaki DT, Murray BW, O'Leary EC, Sakata ST, Xu W, Leisten JC, Motiwala A, Pierce S, Satoh Y, Bhagwat SS, Manning AM, Anderson DW. SP600125, an anthrapyrazolone inhibitor of Jun N-terminal kinase. *Proc Natl Acad Sci USA* 2001; **98**: 13681-13686 [PMID: 11717429 DOI: 10.1073/pnas.251194298]

186 **Grassi ES**, Vezzoli V, Negri I, Lábadi Á, Fugazzola L, Vitale G, Persani L. SP600125 has a remarkable anticancer potential against undifferentiated thyroid cancer through selective action on ROCK and p53 pathways. *Oncotarget* 2015; **6**: 36383-36399 [PMID: 26415230 DOI: 10.18632/oncotarget.5799]

187 **Kim JH**, Kim TH, Kang HS, Ro J, Kim HS, Yoon S. SP600125, an inhibitor of Jnk pathway, reduces viability of relatively resistant cancer cells to doxorubicin. *Biochem Biophys Res Commun* 2009; **387**: 450-455 [PMID: 19607816 DOI: 10.1016/j.bbrc.2009.07.036]

188 **Li JY**, Huang JY, Xing B, Ren KW, Li M, Wei D, Gu PY, Chen G, Gu B, Zhang GF, Hu WX. SP600125, a JNK inhibitor, suppresses growth of JNK-inactive glioblastoma cells through cell-cycle G2/M phase arrest. *Pharmazie* 2012; **67**: 942-946 [PMID: 23210245]

189 **Lu YY**, Chen TS, Wang XP, Qu JL, Chen M. The JNK inhibitor SP600125 enhances dihydroartemisinin-induced apoptosis by accelerating Bax translocation into mitochondria in human lung adenocarcinoma cells. *FEBS Lett* 2010; **584**: 4019-4026 [PMID: 20709060 DOI: 10.1016/j.febslet.2010.08.014]

190 **Lipner MB**, Peng XL, Jin C, Xu Y, Gao Y, East MP, Rashid NU, Moffitt RA, Herrera Loeza SG, Morrison AB, Golitz BT, Vaziri C, Graves LM, Johnson GL, Yeh JJ. Irreversible JNK1-JUN inhibition by JNK-IN-8 sensitizes pancreatic cancer to 5-FU/FOLFOX chemotherapy. *JCI Insight* 2020; **5** [PMID: 32213714 DOI: 10.1172/jci.insight.129905]

191 **Ebelt ND**, Kaoud TS, Edupuganti R, Van Ravenstein S, Dalby KN, Van Den Berg CL. A c-Jun N-terminal kinase inhibitor, JNK-IN-8, sensitizes triple negative breast cancer cells to lapatinib. *Oncotarget* 2017; **8**: 104894-104912 [PMID: 29285221 DOI: 10.18632/oncotarget.20581]

192 **Zhang T**, Inesta-Vaquera F, Niepel M, Zhang J, Ficarro SB, Machleidt T, Xie T, Marto JA, Kim N, Sim T, Laughlin JD, Park H, LoGrasso PV, Patricelli M, Nomanbhoy TK, Sorger PK, Alessi DR, Gray NS. Discovery of potent and selective covalent inhibitors of JNK. *Chem Biol* 2012; **19**: 140-154 [PMID: 22284361 DOI: 10.1016/j.chembiol.2011.11.010]

193 **Cicenas J**, Zalyte E, Rimkus A, Dapkus D, Noreika R, Urbonavicius S. JNK, p38, ERK, and SGK1 Inhibitors in Cancer. *Cancers (Basel)* 2017; **10** [PMID: 29267206 DOI: 10.3390/cancers10010001]

194 **Kumar S**, Jiang MS, Adams JL, Lee JC. Pyridinylimidazole compound SB 203580 inhibits the activity but not the activation of p38 mitogen-activated protein kinase. *Biochem Biophys Res Commun* 1999; **263**: 825-831 [PMID: 10512765 DOI: 10.1006/bbrc.1999.1454]

195 **Lee JC**, Laydon JT, McDonnell PC, Gallagher TF, Kumar S, Green D, McNulty D, Blumenthal MJ, Heys JR, Landvatter SW, Strickler JE, McLaughlin MM, Siemens IR, Fisher SM, Livi GP, White JR, Adams JL, Young PR. A protein kinase involved in the regulation of inflammatory cytokine biosynthesis. *Nature* 1994; **372**: 739-746 [PMID: 7997261 DOI: 10.1038/372739a0]

196 **Habiro A**, Tanno S, Koizumi K, Izawa T, Nakano Y, Osanai M, Mizukami Y, Okumura T, Kohgo Y. Involvement of p38 mitogen-activated protein kinase in gemcitabine-induced apoptosis in human pancreatic cancer cells. *Biochem Biophys Res Commun* 2004; **316**: 71-77 [PMID: 15003513 DOI: 10.1016/j.bbrc.2004.02.017]

197 **Koizumi K**, Tanno S, Nakano Y, Habiro A, Izawa T, Mizukami Y, Okumura T, Kohgo Y. Activation of p38 mitogen-activated protein kinase is necessary for gemcitabine-induced cytotoxicity in human pancreatic cancer cells. *Anticancer Res* 2005; **25**: 3347-3353 [PMID: 16101149]

198 **Pereira L**, Igea A, Canovas B, Dolado I, Nebreda AR. Inhibition of p38 MAPK sensitizes tumour cells to cisplatin-induced apoptosis mediated by reactive oxygen species and JNK. *EMBO Mol Med* 2013; **5**: 1759-1774 [PMID: 24115572 DOI: 10.1002/emmm.201302732]

199 **Paillas S**, Boissière F, Bibeau F, Denouel A, Mollevi C, Causse A, Denis V, Vezzio-Vié N, Marzi L, Cortijo C, Ait-Arsa I, Askari N, Pourquier P, Martineau P, Del Rio M, Gongora C. Targeting the p38 MAPK pathway inhibits irinotecan resistance in colon adenocarcinoma. *Cancer Res* 2011; **71**: 1041-1049 [PMID: 21159664 DOI: 10.1158/0008-5472.CAN-10-2726]

200 **Davies SP**, Reddy H, Caivano M, Cohen P. Specificity and mechanism of action of some commonly used protein kinase inhibitors. *Biochem J* 2000; **351**: 95-105 [PMID: 10998351 DOI: 10.1042/0264-6021:3510095]

201 **Underwood DC**, Osborn RR, Kotzer CJ, Adams JL, Lee JC, Webb EF, Carpenter DC, Bochnowicz S, Thomas HC, Hay DW, Griswold DE. SB 239063, a potent p38 MAP kinase inhibitor, reduces inflammatory cytokine production, airways eosinophil infiltration, and persistence. *J Pharmacol Exp Ther* 2000; **293**: 281-288 [PMID: 10734180]

202 **Rena G**, Bain J, Elliott M, Cohen P. D4476, a cell-permeant inhibitor of CK1, suppresses the site-specific phosphorylation and nuclear exclusion of FOXO1a. *EMBO Rep* 2004; **5**: 60-65 [PMID: 14710188 DOI: 10.1038/sj.embor.7400048]

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