

# Role of p38 and JNK in liver ischemia and reperfusion

LaShonda A. King · Alexander H. Toledo ·  
Fernando A. Rivera-Chavez · Luis H. Toledo-Pereyra

Received: 31 March 2009 / Accepted: 1 July 2009 / Published online: 13 August 2009  
© Springer 2009

## Abstract

**Background/purpose** The signal transduction of mitogen-activated protein kinases (MAPKs) has appeared to be an important mediator of ischemic-related events. Because of this, we analyzed the participation of p38 and JNK in liver ischemia and reperfusion, as two individual members of the MAPK family of proteins.

**Methods** All papers referred to in PubMed for the past 15 years were analyzed to determine how and when these MAPKs were considered to be an intricate part of the ischemic event. References were cross-studied to ascertain whether other papers could be found in the literature.

**Results** The role of p38 and JNK in liver ischemia was confirmed in the literature. The activation of these mediators was associated with the induction of apoptosis and necrosis. Inhibitors of p38 and JNK reduced the liver ischemia and reperfusion damage, probably through the mechanisms mentioned before.

**Conclusions** The development of effective inhibitors of p38 and JNK protein mediators is important for minimizing the harmful effects associated with liver ischemia and reperfusion.

**Keywords** Liver ischemia · Reperfusion · p38 · JNK · Ischemia mediators

## Introduction

Ischemia and reperfusion (I/R) injury represents a complex series of events that result in cellular and tissue damage. It is the transient deprivation and return of blood flow and oxygen that produces a concomitant release of oxygen-free radicals, cytokines/chemokines, and upregulation of adhesion molecules. The consequence is cellular and organ dysfunction [1]. Liver I/R injury causes up to 10% of early organ dysfunction after transplantation, leading to a high incidence of episodes of both acute and chronic rejection. This is because liver I/R injury results not only in hepatocyte necrosis but also in apoptosis. Features of apoptosis and necrosis often coexist, a phenomenon termed necroptosis [2]. I/R injury also contributes to the shortage of organs available for transplantation because of the greater susceptibility of donor livers to ischemic insult.

At present there is no specific treatment available to prevent organ damage caused by liver I/R injury. Thus, injuries due to I/R remain an important source of morbidity and mortality in surgery, trauma, and transplantation [3]. Given the need for effective I/R injury treatment, much research has been done on the mechanisms of I/R injury and the development of potential therapeutic strategies [1, 4].

One mechanism of interest is the mitogen-activated protein kinase (MAPK) family, which plays an important role in intracellular signal transduction in response to extracellular stimuli. Among mammalian MAPKs, p38 and c-Jun N-terminal kinase (JNK) are activated by a variety of cellular stresses, such as I/R [5–16]. This review will focus

---

L. A. King · L. H. Toledo-Pereyra (✉)  
Departments of Research and Surgery,  
Kalamazoo Center for Medical Studies,  
Michigan State University, 1000 Oakland Drive,  
Kalamazoo, MI 49008, USA  
e-mail: Toledo@kcms.msu.edu

A. H. Toledo  
Division of Abdominal Transplantation,  
University of North Carolina, Chapel Hill, NC, USA

F. A. Rivera-Chavez  
University of Texas Southwestern Medical Center,  
Dallas, TX, USA

on the role of p38 and JNK in liver I/R injury and the related therapeutic implications.

### MAPKs contribute to liver I/R

The three well-characterized subfamilies of MAPKs are extracellular signal regulated kinases (ERKs), c-Jun N-terminal kinases (JNKs), and p38 [12]. These kinases are 60–70% identical to each other. They differ in the sequence and size of their activation loop, as well as in their activation in response to different stimuli. In general, ERKs are activated by mitogenic and proliferative stimuli, whereas JNKs and p38s respond to environmental stress, and are referred to as stress-activated protein kinases (SAPKs) [12, 13]. Upon stimulation, these kinases are activated by the dual phosphorylation of threonine (Thr) and tyrosine (Tyr) residues. MAPKs are phosphorylated and activated by MAPK-kinase (MAPKK) whereas MAPKKs are activated through phosphorylation by MAPKK-kinases (MAPKKKs) [17]. The MAPK pathways are not independent from each other but contain a series of overlapping signaling mechanisms (Fig. 1). Thus, p38, JNK, and ERK have interrelated functions [8]. Once activated, these kinases are translocated to the nucleus, where they phosphorylate and activate different transcription factors and transactivate target genes [14]. For example, JNK and p38 both phosphorylate and activate activating transcription factor 2 (ATF-2) [12].

There is much evidence demonstrating the role of MAPKs, specifically p38 and JNK, in damage following liver I/R. p38 and JNK are phosphorylated and activated several minutes after reperfusion [5, 12, 15]. Their activation is associated with the induction of apoptosis and necrosis [2, 5]. Treatment with p38/JNK activators increased transaminase levels and grade 3 necrosis in rat hepatic tissue sections. However, MAPK inhibition reduced hepatic I/R injury in the liver [16].

p38 and JNK also contribute to liver fibrosis [17]. A recent study showed that basal JNK and p38 kinase activities were higher in activated hepatic stellate cells (HSCs) than in quiescent HSCs. HSCs are responsible for the synthesis of type I collagen after liver injury and often cause liver fibrosis as a result of their activation. Mediators of inflammation (eicosanoids, tumor necrosis factor [TNF]- $\alpha$ , interleukin [IL]-1, adhesion molecules, NO, and intracellular  $\text{Ca}^{2+}$  [ $\text{iCa}^{2+}$ ]) begin a series of cascades associated with the damage characteristics of I/R. MAPKs are central to this pathologic event, with their signaling and gene response dependent on the degree of severity of I/R. Manipulation of these molecular inflammatory cascades with MAPK inhibitors offers a possibility for overcoming inflammatory damage caused by I/R [8, 18].

### p38 and JNK are activated during ischemic preconditioning

MAPKs can also provide a protective feature through the use of ischemic preconditioning. Ischemic preconditioning is a technique whereby an organ is rendered resistant to the damaging effects of I/R by prior exposure to a brief period of vascular occlusion [19]. Ischemic preconditioning has been successful for tumor hepatic resections under normothermic conditions [16].

There is *in vivo* evidence in isolated hepatocytes and experimental rat models of perfused livers that MAPKs are involved in this protective mechanism [16]. For example, ischemic preconditioning activates p38 and JNK-1. This activation is associated with increased cyclin D1 expression and entry into the cell cycle [20]. Induction of cyclin D1 is one of the earliest and most pivotal steps in the pathway of resting cells to enter the cell cycle. p38 and JNK both control cyclin D1 expression; however, the protective effect of the preconditioning regimen was more closely associated with p38 than with JNK-1 activation [19].

### p38 and liver I/R

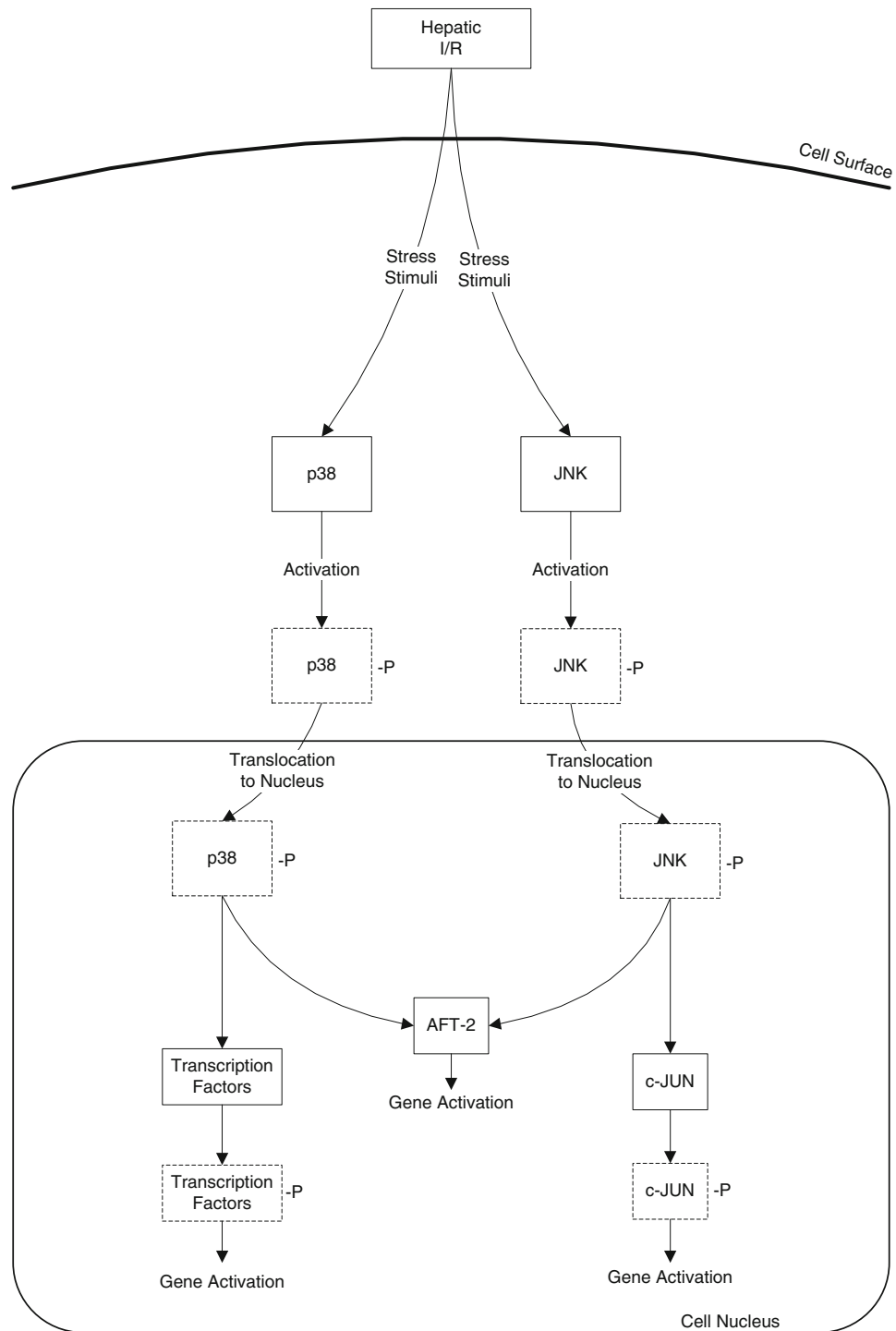
p38 was discovered in three independent contexts: first as a tyrosine phosphoprotein in extracts of cells treated with inflammatory cytokines; second, as a target of a pyridyl imidazole drug that blocks the production of TNF- $\alpha$ ; and third, as a reactivating kinase for MAP kinase-activated protein (MAPKAP) [21]. There are four isoforms of the protein: p38 $\alpha$ , p38 $\beta$ , p38 $\delta$ , and p38 $\gamma$  [7]. The  $\beta$ ,  $\delta$ , and  $\gamma$  isoforms share sequence homology with the  $\alpha$  isoform (74, 57, and 60%, respectively) [7].

p38 proteins are activated by cytokines, hormones, G protein-coupled receptors (GPCRs), osmotic shock, heat shock, and other stresses [6]. p38 is activated by dual phosphorylation on Thr180 and Try182 by an upstream MAPKK termed MAP2K6 (also known as MKK6). Other MAPKKs may also activate p38. The p38 isoforms phosphorylate a wide array of targets, thus inducing other kinases [7, 8]. Many of the downstream targets of activated p38 are responsible for the transactivation of genes encoding inflammatory cytokines [13].

The main biological response of p38 activation involves the production and activation of inflammatory mediators to initiate leukocyte recruitment and activation [22]. Activation of the p38 pathway and p38-associated inflammatory processes play a crucial role in post-ischemic damage in the liver [7].

Recent research has focused on potential inhibitors of p38 and their therapeutic implications (Tables 1, 2). p38 inhibitors block the production and activity of

**Fig. 1** Overlapping signaling mechanisms of p38 and c-Jun N-terminal kinase (*JNK*) pathways. Environmental stresses induce the activation of p38 and JNK. Once activated p38 and JNK are translocated to the nucleus where they phosphorylate and activate transcription factors and genes. The pathways contain overlapping signaling mechanisms



inflammatory cytokines [13]. Inhibition of p38 activity prevents the transactivation of both the IL-1 $\alpha$  and IL-1 $\beta$  genes which encode IL-1 in lipopolysaccharide (LPS)-stimulated macrophages in vitro [23]. In rat models, the addition of a p38 inhibitor blocks the activation of p38 after reperfusion, decreases reperfusion injury in the liver graft, and increases the survival of recipients [15] (Table 2; Fig. 2).

Inhibitors targeting the p38 pathway have been developed, and preliminary preclinical data suggests they exhibit anti-inflammatory activity. The ability of p38 inhibitors to block TNF- $\alpha$  synthesis is being exploited in the treatment of inflammatory diseases, and p38 inhibitors have been shown to inhibit the development of rheumatoid arthritis in small animal models [23]. Thus, the use of MAPK inhibitors has emerged as an attractive strategy because they are

**Table 1** Summary of experimental p38 results after warm/cold ischemia of the liver

Experiment	Results	Reference
I/R in pigs given ANP	AST, LDH, and tissue damage decreased	[26]
Liver transplantations performed in rats	Sustained induction of JNK but not p38	[14]
I/R in rats given ANP preconditioning	Activates p38, alters hepatocyte cytoskeleton	[24]
Total I/R in rats	p38 highly phosphorylated	[12]
Rat liver transplantation with p38 inhibitor FR167653	Lowers ALT and LDH levels, improves survival	[15]
I/R in rats given ANP	Increased p38 activity	[25]
I/R in mice with ischemic preconditioning	Protective effect of ischemic preconditioning associated with p38	[19]
I/R in animals given p38/JNK activators	Increased transaminase levels and grade 3 necrosis	[16]
I/R in mice given ischemic preconditioning and TNF- $\alpha$	Preconditioning and TNF- $\alpha$ pretreatments both give hepatoprotection	[20]
Hepatic stellate cells (HSCs) exposed to SP600125, a JNK inhibitor, and SB203580, a p38 inhibitor	p38 inhibition increased and JNK inhibition decreased HSC proliferation	[17]
I/R and caspase inhibitor in rats	p38 and JNK have different patterns of activation	[2]

I/R ischemia and reperfusion, ANP atrial natriuretic peptide, AST aspartate aminotransferase, LDH lactate dehydrogenase, JNK c-Jun N-terminal kinase, ALT alanine aminotransferase, TNF- $\alpha$  tumor necrosis factor- $\alpha$

**Table 2** p38 inhibitors utilized in liver I/R injury

Inhibitor	Effect	Reference
SB203580	Increased proliferation rate of HSCs	[17]
SB203580	Inhibits TNF- $\alpha$ and cytokine synthesis	[22]
FR167653	Increased survival, decreased serum ALT and LDH	[15]
SB202190	Inhibits programmed cell death	[30]
SKF-86002	Blocks production of cytokines	[13]
SB220025	Inhibits TNF- $\alpha$	[22]

All the compounds presented above decreased liver I/R injury, except for SB203580

capable of reducing both the synthesis of pro-inflammatory cytokines and their signaling [22].

In addition to cytokine activation, damage to liver tissue following I/R injury is also associated with cytoskeletal changes. In liver cells F-actin forms microfilaments, which are involved in intracellular transport processes such as exocytosis and endocytosis, maintenance of cell shape, and canalicular motility responsible for bile flow. Structural alterations of the cytoskeleton have been reported to cause disturbances of intracellular transport processes, cell motility, and microcirculation following I/R, leading to dysfunction of the organ. After ischemia in vivo, F-actin is reduced in rabbit livers, resulting in the loss of cell integrity and cytoplasmic transport in the liver, causing damage to organelles and changes in cell morphology. It has been shown experimentally that atrial natriuretic peptide (ANP)

leads to the activation of p38. p38 activation leads to hepatocyte cytoskeletal changes by increasing the hepatocyte F-actin content in rat livers [24].

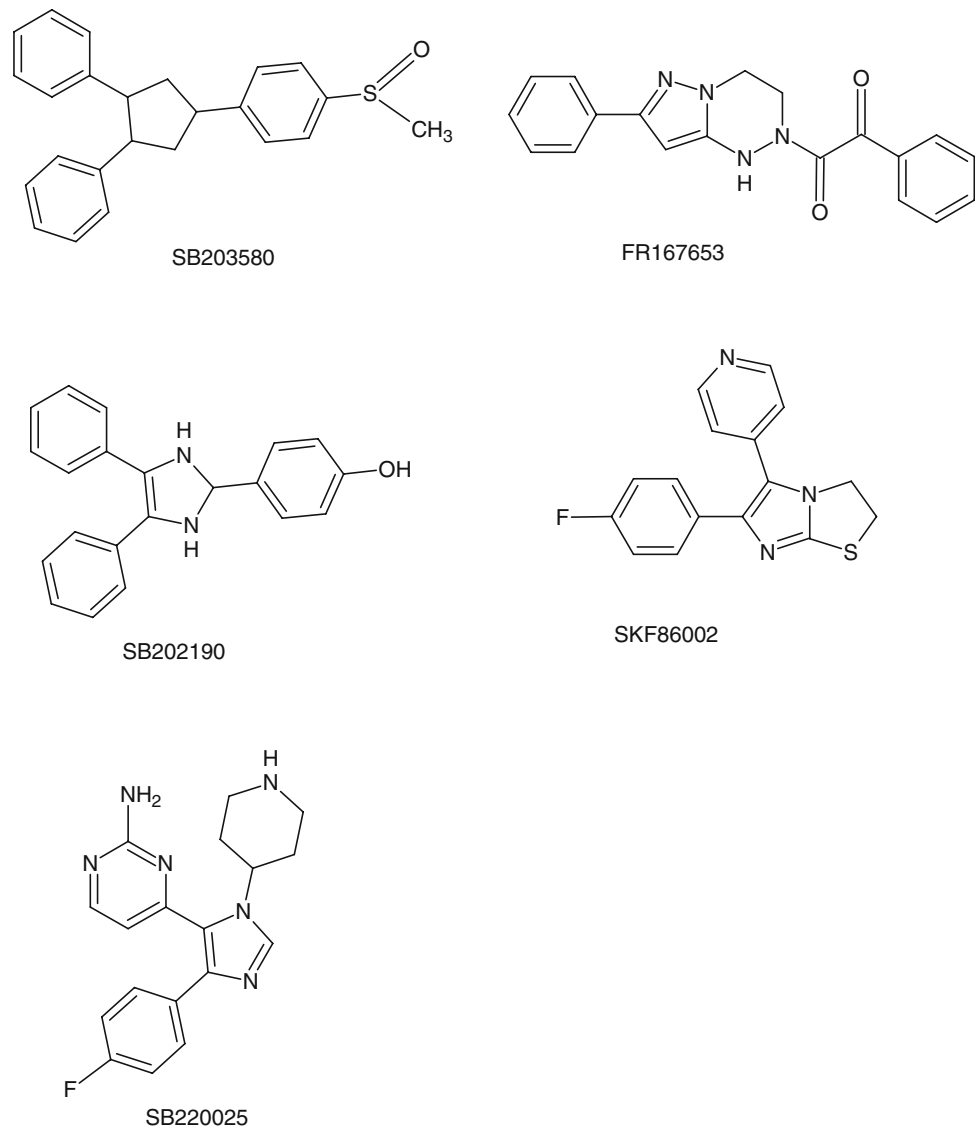
Preconditioning with ANP increases p38 activity in the liver in vivo [24]. The activation of p38 at the end of the preconditioning period (i.e., before ischemia) is thought to be the crucial step in mediating hepatoprotection by ANP [25]. Following reperfusion in the rat model, phosphorylated p38, the active form of p38, was higher in the ANP-treated group than in the control group. Histological tissue damage was milder in the ANP group than in the control group [26]. Thus, p38 has a dual role, it serves as a protectant stimulating the production of factors such as ANP which protect against damage caused by I/R, yet it also acts as a catalyst for liver damage.

### JNK and liver I/R

JNK is also a member of the MAP kinase family. JNK is a 54-kDa Ser/Thr kinase that is activated in response to several conditions including heat shock, ultraviolet radiation, protein synthesis inhibitors, inflammatory cytokines such as IL-1 and TNF, oxidative stress, and LPS [8, 27]. Three JNK isoforms have been identified in humans: JNK1, JNK2, and JNK3 [9]. JNK1 and JNK2 exist in the liver [10].

In 1994, activated JNK was observed in I/R injury in the heart and kidneys [11]. Since this initial discovery, JNK activation in I/R injury has been observed in other organs such as the liver. It has been shown experimentally that JNK is activated by TNF- $\alpha$  and IL-1 cytokines. These cytokines are major mediators of hepatic I/R injury. JNK also mediates inflammatory processes by inducing the

**Fig. 2** Structures of p38 inhibitors utilized in experimental liver I/R injury



expression of adhesion molecules and inflammatory chemokines [28].

JNK activation is also associated with hepatocyte apoptosis. The death signaling pathway which leads to apoptosis includes caspase 3 activation and the release of cytochrome c. Experimental inhibition of JNK with specific selective inhibitors such as CC-401 decreases hepatocyte apoptosis in rats in vivo following orthotopic liver transplantation by decreasing cytochrome c release and caspase 3 activation [29] (Tables 3, 4; Fig. 3).

High oxidative stress environments, such as those demonstrated during I/R, also lead to JNK activation and elevated reactive oxygen species (ROS) production. Under I/R conditions, JNK activation is initiated in the mitochondria and requires coupled electron transport, ROS generation, and calcium flux [7]. Consequently, the modulation of JNK activation by oxygen-free radical inhibitors

might offer a new way of preventing the consequences of I/R of the liver [30].

Numerous studies have shown that inhibition of JNK prevents damage associated with I/R. For example, JNK inhibition significantly decreased nonparenchymal cell death 60 min after reperfusion and pericentral necrosis 8 h after reperfusion. JNK inhibition decreases hepatic necrosis and apoptosis after orthotopic liver transplantation. JNK inhibition also enhances graft survival, decreases graft injury, and improves liver function after orthotopic liver transplantation. JNK inhibition did not affect other signaling pathways, such as p-38 and Erk activation [29].

JNK inhibition suppresses liver injury in a rat model of hepatic warm I/R [31]. JNK induces mitochondrial damage with the release of cytochrome c into the cytoplasm. This release of cytochrome c is an integral component of the mitochondrial pathway of apoptosis. The study also

**Table 3** Summary of experimental JNK results after warm/cold ischemia of the liver

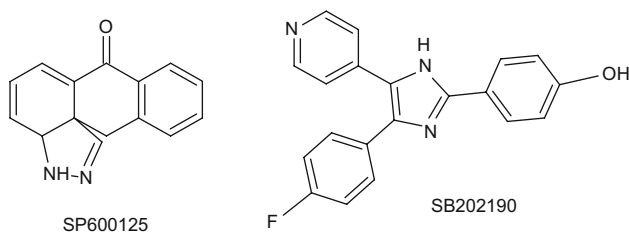
Experiment	Results	Reference
Warm and cold ischemia in rat hepatocytes	JNK activity and apoptosis induced	[30]
I/R of rats followed by partial hepatectomy	JNK inhibitor causes decreased apoptosis/necrosis, increased survival	[31]
Partial liver I/R in rats	Reperfusion rapidly activates JNKs and ERKs, marked activation of JNKs	[27]
Treated liver explants with JNK inhibitor before OLT with cold storage	Decreased cell killing, pericentral necrosis, cytochrome c release, caspase 3 activation, lipid peroxidation	[29]
Partial I/R in mice given SP600125 JNK inhibitor	Increase in ALT levels, parenchymal destruction, leukocyte infiltration	[28]
Rat hepatocytes given SP600125 JNK inhibitor and exposed to apoptotic stimuli	Improved viability, decreased DNA nick end labeling positivity, cleavage of ADP-ribose polymerase and caspase-3	[32]
Total hepatic ischemia in mice	JNK not activated during ischemia. Activity increased during reperfusion	[11]
Warm I/R in rats given melatonin	Improved survival and decreased transaminase levels, JNK1, and cJUN	[6]

ERK extracellular signal regulated kinase, OLT orthotopic liver transplantation

**Table 4** JNK inhibitors utilized in liver I/R injury

Inhibitor	Effect	Reference
CC-401	Decreases release of AST and tissue TNF- $\alpha$ Decreases necrosis, apoptosis, cytochrome c release, caspase 3 activation	[29, 31]
SB202190	Inhibits programmed cell death	[30]
CC0209766	Decreases necrosis, apoptosis, cytochrome c release, caspase 3 activation	[31]
CC0223105	Decreases necrosis, apoptosis, cytochrome c release, caspase 3 activation	[31]
SP600125	Aggravates reperfusion injury, increases ALT, leukocyte infiltration, parenchymal destruction, decreases HSC proliferation	[17, 28]

All of the compounds presented above decreased liver I/R injury, except for SP600125



**Fig. 3** Structures of JNK inhibitors utilized in experimental liver I/R injury. The other three JNK inhibitors presented in Table 4 are not drawn in this figure because of intellectual property issues

showed that JNK inhibitors decrease tissue levels of TNF- $\alpha$  and decrease lipid peroxidation. In vitro, JNK inhibitors block c-Jun phosphorylation, expression of inflammatory genes, and cell proliferation. These data indicate that JNK inhibitors decrease both hepatic necrosis and apoptosis caused by hepatic warm I/R [31].

Many of the studies evaluating the role of JNK activation in apoptosis have relied on knockout animals or viral gene delivery systems. The development of the pharmacological adenosine triphosphate-competitive JNK inhibitor SP600125 offered a simple and direct technique to study this role. Pharmacological inhibition of JNK

activation protects rat hepatocytes from apoptosis and improves cell viability. Pretreatment with JNK inhibitor significantly protected against cell death associated with apoptotic stimuli such as TNF- $\alpha$  and actinomycin D. This protection was associated with maintaining the expression of anti-apoptotic proteins [32].

The above discussion of the role of JNK in damage due to hepatic I/R highlights the possibility and benefit of using JNK inhibitors clinically. Inhibition of JNK may be a new therapeutic strategy to prevent liver injury after transplantation [29]. Likewise, antioxidants and JNK inhibitors appear to be useful drugs for the treatment of TNF-dependent hepatitis. There is also potential use for inhibitors in the treatment of fulminant hepatitis and hepatocellular carcinoma in clinical settings [10].

Attention has also been given to JNK as a potential target for the development of novel therapeutics. Inhibitors targeting JNK pathways have been developed and preliminary preclinical data suggest that they exhibit anti-inflammatory activity [23]. JNK mediates the phosphorylation and activation of ATF2, which contributes to the induction of TNF- $\alpha$  and interferon  $\beta$ . JNK inhibitors have been tested in rat models of rheumatoid arthritis and found to be effective in reducing inflammation [23].

Melatonin has also emerged as a potential treatment option for liver I/R injury through its inhibition of JNK. Experiments have shown that melatonin displays the same characteristics of inhibiting JNK and cJUN as other pharmacological inhibitors of JNK which reduce hepatocyte necrosis and improve survival after warm I/R and liver resection. Thus, considering the low potential of harmful side effects, melatonin is an attractive modality for treating liver I/R injury [6].

## Conclusion

p38 and JNK, which are induced by extracellular stresses, contribute to liver injury caused by I/R. Discovery of a treatment to regulate these kinases and their activities will be beneficial in the management and prevention of liver damage. p38 and JNK inhibitors have great potential therapeutic value. These inhibitors reduce cellular damage, apoptosis, and necrosis following liver I/R.

There are, however, limitations to the use of p38 and JNK inhibitors that merit more investigation. In this regard, p38 offers protection from damage during ischemic preconditioning. Additionally, inhibition of p38 has been found to increase the proliferation of HSCs, which can lead to liver fibrosis [17]. Furthermore, in another study, JNK inhibition aggravated I/R injury and caused more severe parenchymal destruction [28]. The study was evaluating the use of the SP600125 JNK inhibitor in hepatic I/R injury using a partial ischemia model in mice. The data, although contradictory to other studies, confirm the need for more investigation into the use of JNK inhibitors [28].

More research is needed to translate p38 and JNK inhibitors from the laboratory to clinical application. A more extensive definition of the mechanisms by which these kinases work would greatly improve the development and use of these inhibitors. Better understanding of the pathways and molecular structure of p38 and JNK inhibitors would permit the inhibition of only the portions that are necessary to prevent liver I/R injury.

## References

- Vardanian AJ, Busuttil RW, Kupiec-Weglinski JW. Molecular mediators of liver ischemia and reperfusion injury: a brief review. *Mol Med*. 2008;14:337–45.
- Cursio R, Filippa N, Miele C, Van Obberghen E, Gugenheim J. Involvement of protein kinase B and mitogen-activated protein kinases in experimental normothermic liver ischemia–reperfusion injury. *Brit J Surg*. 2006;93:752–61.
- Lopez-Neblina F, Toledo AH, Toledo-Pereyra LH. Molecular biology of apoptosis in ischemia and reperfusion. *J Invest Surg*. 2005;18:335–50.
- Busuttil R. Liver ischemia and reperfusion injury. *Brit J Surg*. 2007;94:787–8.
- Kobayashi M, Takeyoshi I, Yoshinari D, Matsumoto K, Morishita Y. p38 mitogen-activated protein kinase inhibition attenuates ischemia–reperfusion injury of the rat liver. *Surgery*. 2002;131:344–9.
- Liang R, Nickkholgh A, Hoffmann K, Kern M, Schneider H, Sobirey M, et al. Melatonin protects from hepatic reperfusion injury through inhibition of IKK and JNK pathways and modification of cell proliferation. *J Pineal Res*. 2009;46:8–14.
- Toledo-Pereyra LH, Lopez-Neblina F, Toledo AH. Protein kinases in organ ischemia and reperfusion. *J Invest Surg*. 2008;21:215–26.
- Toledo-Pereyra LH, Toledo AH, Walsh J, Lopez-Neblina F. Molecular signaling pathways in ischemia/reperfusion. *Exp Clin Transplant*. 2004;2:174–7.
- Tsung A, Hoffman RA, Izuishi K, Critchlow ND, Nakao A, Chan MH, et al. Hepatic ischemia/reperfusion injury involves functional TLR4 signaling in nonparenchymal cells. *J Immunol*. 2005;175:7661–8.
- Schwabe RF, Brenner DA. Mechanisms of liver injury. I. TNF- $\alpha$ -induced liver injury: role of IKK, JNK, and ROS pathways. *Am J Physiol Gastrointest Liver Physiol*. 2006;290:G583–9.
- Shimizu OK, Tani T, Hashimoto T, Miwa K. JNK activation and apoptosis during ischemia–reperfusion. *Transplant Proc*. 1999;31:1077–9.
- Kobayashi M, Takeyoshi I, Yoshinari D, Matsumoto K, Morishita Y. The role of mitogen-activated protein kinases and the participation of intestinal congestion in total hepatic ischemia–reperfusion injury. *Hepatogastroenterology*. 2006;53:243–8.
- Kumar S, Boehm J, Lee JC. p38 MAP Kinases: key signaling molecules as therapeutic targets for inflammatory diseases. *Nat Rev Drug Discov*. 2003;2:717–25.
- Bradham CA, Stachlewitz RF, Gao W, Qian T, Jayadev S, Jenkins G, et al. Reperfusion after liver transplantation in rats differentially activates the mitogen-activated protein kinases. *Hepatology*. 1997;25:1128–35.
- Yoshinari D, Takeyoshi I, Kobayashi M, Koyama T, Iijima K, Ohwada S, et al. Effects of a p38 mitogen-activated protein kinase inhibitor as an additive to University of Wisconsin solution on reperfusion injury in liver transplantation. *Transplantation*. 2001;72:22–7.
- Massip-Salcedo M, Casillas-Ramirez A, Franco-Gou R, Bartrons R, Mosbah IB, Serafin A, et al. Heat shock proteins and mitogen-activated protein kinases in steatotic livers undergoing ischemia–reperfusion: some answers. *Am J Pathol*. 2006;168:1474–85.
- Schnabl B, Bradham CA, Bennett BL, Manning AM, Stefanovic B, Brenner DA. TAK1/JNK and p38 have opposite effects on rat hepatic stellate cells. *Hepatology*. 2001;34:953–63.
- Lopez-Neblina F, Toledo-Pereyra LH. Phosphoregulation of signal transduction pathways in ischemia and reperfusion. *J Surg Res*. 2006;134:292–9.
- Teoh N, Pena AD, Farrell G. Hepatic ischemic preconditioning in mice is associated with activation of NF- $\kappa$ B, p38 kinase, and cell cycle entry. *Hepatology*. 2002;36:94–102.
- Teoh N, Laclercq I, Pena AD, Farrell G. Low dose TNF- $\alpha$  protects against hepatic ischemia–reperfusion injury in mice: implications for preconditioning. *Hepatology*. 2003;37:118–28.
- Interpro Database, European Bioinformatics Institute (2006–2009) <http://www.ebi.ac.uk/interpro/Entry?ac=IPR008352>.
- Kaminska B. MAPK signaling pathways as molecular targets for anti-inflammatory therapy—from molecular mechanisms to therapeutic benefits. *Biochem Biophys Acta*. 2005;1754:253–62.

23. Karin M. Mitogen activated protein kinases as targets for development of novel anti-inflammatory drugs. *Ann Rheum Dis.* 2004;63:ii62–4.
24. Keller M, Gerbes AL, Kulhanek-Heinze S, Gerwig T, Grutzner U, Van Rooijen N, et al. Hepatocyte cytoskeleton during ischemia and reperfusion—influence of ANP-mediated p38 MAPK activation. *World J Gastroenterol.* 2005;11:7418–29.
25. Kiemer AK, Kulhanek-Heinze S, Gerwig T, Gerbes AL, Vollmar AM. Stimulation of p38 MAPK by hormonal preconditioning with atrial natriuretic peptide. *World J Gastroenterol.* 2002;8:707–11.
26. Kobayashi K, Oshima K, Muraoka M, Akao T, Totsuka O, Shimizu H, et al. Effect of atrial natriuretic peptide on ischemia–reperfusion injury in a porcine total hepatic vascular exclusion model. *World J Gastroenterol.* 2007;13:3487–92.
27. Bendinelli P, Piccoletti R, Maroni P, Bernelli-Zazzera A. The MAP kinase cascades are activated during post-ischemic liver reperfusion. *FEBS Lett.* 1996;398:193–7.
28. Lee KH, Kim SE, Lee YS. SP600125, a selective JNK inhibitor, aggravates hepatic ischemia–reperfusion injury. *Exp Mol Med.* 2006;38:408–16.
29. Uehara T, Xi Peng X, Bennett B, Satoh Y, Friedman G, Currin R, et al. c-Jun N-terminal kinase mediates hepatic injury after rat liver transplantation. *Transplantation.* 2004;78:324–32.
30. Crenesse D, Gugenheim J, Hornoy J, Tornieri K, Laurens M, Cambien B, et al. Protein kinase activation by warm and cold hypoxia-regeneration in primary-cultured rat hepatocytes—JNK1/SAPK1 involvement in apoptosis. *Hepatology.* 2000;32:1029–36.
31. Uehara T, Bennett B, Sakata ST, Satoh Y, Bilter GK, Westwick JK, et al. JNK mediates hepatic ischemia reperfusion injury. *J Hepatol.* 2005;42:850–9.
32. Marderstein EL, Bucher B, Zhong Guo BS, Feng X, Reid K, Geller DA. Protection of rat hepatocytes from apoptosis by inhibition of c-Jun N-terminal kinase. *Surgery.* 2003;134:280–4.