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Chapter

Hepatic Regeneration Under Warm or Cold Ischemia Conditions: Controversies and New Approaches

Maria Eugenia Cornide-Petronio, Mónica B. Jiménez-Castro, Esther Bujaldon, Jordi Gracia-Sancho and Carmen Peralta

Abstract

Ischemia-reperfusion (I/R) associated with hepatic resection and living related liver transplantation is an unsolved problem in clinical practice. Indeed, I/R induces damage and regenerative failure in clinical liver surgery. Signaling pathways regarding the pathophysiology of liver I/R and regeneration making clear distinction between situations of cold and warm ischemia, as well as liver regeneration with or without vascular occlusion, will be addressed. The different experimental models used to date to improve the postoperative outcomes in clinical liver surgery will be also described. Furthermore, the most updated therapeutic strategies, as well as the clinical and scientific controversies in the field, will be discussed. Such information may be useful to guide the design of better experimental models as well as the effective therapeutic strategies in liver surgery that can succeed in achieving its clinical application.

Keywords: liver surgery, regeneration, ischemia–reperfusion injury, warm ischemia, cold ischemia

1. Introduction

Any surgical situation involving liver hepatectomy requires subsequent regeneration in order to restore the liver/body ratio. The liver's ability to restore tissue after loss depends on the interaction of numerous cells and a complex network of mediators [1]. In most cases, in clinical practice, liver surgery involves both ischemia-reperfusion (I/R) injury and regeneration [1]. Liver I/R injury is a pathophysiological event that occurs during surgical interventions such as liver resection or liver transplantation (LT); it controls bleeding during parenchymal dissection and has a significant effect on liver function prognosis [2–6]. I/R injury is a two-stage phenomenon in which cell damage due to hypoxia and the lack of biomechanical stimulus is exacerbated upon the restoration of oxygen delivery and shear stress [7]. However, I/R injury is inevitable in liver surgery and significantly reduces the organ's regeneration after hepatectomy [1]. Mechanisms of liver I/R injury are complex; they include mainly microcirculation failure and the related oxidative stress, a series of cellular and molecular responses, and the interaction between hepatocytes,

liver sinusoidal endothelial cells, hepatic stellate cells, Kupffer cells, infiltrating neutrophils, macrophages, and platelets [2, 7–11].

Liver I/R injury involves great many factors and mediators. The associations between the signaling pathways are extremely complex, and at present, the events occurring between the start of reperfusion and the final outcome (either poor function or a nonfunctional liver graft) are not fully understood [6]. The extent and timing of ischemia, the type of liver undergoing I/R, and the existence of liver regeneration may alter the mechanisms of liver I/R injury and the effects of the treatment strategies assessed to date [12]. This point was exemplified by Ramalho et al. who demonstrated the loss of protection against liver damage of Ang-II receptor antagonists in conditions of partial hepatectomy (PH), while in conditions of I/R without hepatectomy, the Ang-II receptor antagonists decreased liver damage [13]. As is well known, the mechanisms of liver damage differ according to the percentage of the liver mass deprived of blood [14]. Therefore, experimental models that reproduce as closely as possible the clinical conditions in which these strategies are applied are likely to lead to the implementation of strategies in clinical practice in the relatively short term [1].

In this chapter, we aim to show that the mechanisms that govern liver I/R injury and regeneration depend on the experimental model applied. These models are valuable tools for elucidating the physiopathology of liver I/R injury and uncovering new therapeutic targets and drugs. A number of strategies for protecting the liver from I/R injury and for improving liver regeneration have been developed in animal models, some of which may find their way into clinical practice [6]. We stress that the type of ischemia (cold or warm) has an important influence in liver surgery, but most of the currently available reviews on the mechanisms of I/R do not distinguish between them [6]. In our view, this information may help to guide the design of future experimental models and treatment strategies in liver surgery for use in clinical practice.

2. Hepatic regeneration under warm ischemia

Following PH, hepatocytes that are normally quiescent enter the cell cycle in order to replace the part that has been removed. The original liver mass is restored after some 6–8 weeks (in humans) by tightly synchronized rounds of replication of the remaining hepatocytes [15]. Self-replication of the existing individual cell types is thought to be the key mechanism of regeneration after PH. However, it was recently suggested that hepatic progenitor cells may contribute to liver regeneration following PH [15]. A vast number of growth and metabolic factors and cytokines simultaneously regulate liver regeneration during PH. Under the influence of innate immunity components and gut-derived lipopolysaccharide, on Kupffer cells and stellate cells, tumor necrosis factor alpha, and interleukin 6, provided by those cells, prepares hepatocytes to respond to growth factors like epidermal growth factor and hepatocyte growth factor [15]. Among other auxiliary mitogens are norepinephrine, Notch and jagged proteins, vascular endothelial growth factor, platelet-derived growth factor, bile acids, insulin serotonin, estrogens leptin, triodothronine, and FGF1 and 2 [16]. Joint signals from these factors lead to the progression of the liver cell cycle, which in turn results in DNA synthesis and ultimately the proliferation of liver cells as mentioned above [15].

As is well known, remnant liver following PH can be used as an in vivo liver regeneration model in order to assess possible treatment strategies for improving postoperative outcomes after hepatectomy. Nonetheless, a two-third partial hepatectomy alone does not cause death in these models, and the remnant liver has the capacity to regenerate. In contrast, 30 min of liver ischemia just before PH exacerbates the remnant liver function, causing high mortality and negatively affecting liver weight restoration [1, 17]. It is also well known that vascular occlusion of the

hepatic hilum is often used to avoid hemorrhage in liver resection. However, vascular occlusion has been associated with warm ischemia damage, resulting in significant organ dysfunction and regenerative failure [12]. Hepatocytes are severely affected by I/R, especially in normothermic ischemia. Most of the early changes in anoxic hepatocytes take place in the mitochondria. Briefly, due to the unavailability of O_2 as a terminal electron carrier for the mitochondrial respiratory chain, the electron flow is immediately interrupted, thus reducing the respiratory chain. Since the mitochondria no longer accept electrons from substrates, pyridine nucleotides decrease, thus causing a rise in the intracellular NADH/NAD^+ ratio. The disruption of oxidative phosphorylation rapidly depletes cellular ATP, accelerates glycolysis, increases lactate formation, and alters H^+, Na^+, and Ca^{2+} homeostasis and thus induces severe damage to the hepatocyte. Ischemia also causes a substantial rise in cAMP, a key factor in glucose metabolism. Via the action of cAMP-dependent protein kinase, cAMP causes the phosphorylation/deregulation of enzymes, which play a major role in the control of carbohydrate metabolism [18, 19]. Reperfusion injury derives mainly from toxic reactive oxygen species (ROS) generated on the reintroduction of O_2 to ischemic tissues. ROS are produced both from intracellular and from extracellular sources; in liver cells, the mitochondria are their major source [7, 20].

3. Preclinical studies in normothermic hepatic ischemia associated with hepatic resection

3.1 Animal species used

The results of animal studies can be extrapolated to human beings, even though there are limitations such as the differences in ischemia tolerance, the anatomy of the organ in different species, and differences in the surgical conditions applied in clinical practice and in experimental models [21]. Therefore, the correct choice of animal species and experimental model, and the standardization of the protocol according to the clinical issue under study, is particularly important [14]. Small animals like rats and mice are exceptionally useful because they are easy to handle, present minimal, financial, logistical or ethical problems, and allow genetic alterations such as the creation of transgenic and knock-out animals [14]. Larger animals (pigs, sheep, and dogs) have a more similar anatomy and physiology to humans, but their use is restricted by serious financial and logistical difficulties, ethical concerns, and the limited availability of immunological tools for use in these species [14, 21]. The age and sex of animals are also issues to consider. With regard to age, there are significant differences between younger (35–50 g) and older rats (250–400 g) in terms of their hepatic microcirculation at the different stages of ischemia, and with regard to sex, female rats are more sensitive to reperfusion injury than males after normothermic ischemia [14, 22, 23].

3.2 Experimental models of normothermic hepatic ischemia to evaluate the mechanisms involved

3.2.1 Global hepatic ischemia with portocaval decompression

The Pringle maneuver is often applied during liver resection, due to safety concerns. However, it has been associated with delayed liver failure and poor prognosis in patients undergoing major hepatectomy in conditions of prolonged liver ischemia [1, 13]. The global liver ischemia model with portal decompression provides an ideal simulation of the clinical condition of warm ischemia after the Pringle maneuver for liver resection and transplant [6, 14]. The first successful

shunt operation carried out in humans was by Vidal in 1903 [24]. Blakemore was among the first researchers to report successful portal-systemic anastomosis in rats, working mainly with endothelium-lined tubes [25]. Burnett et al. modified the technique to create a portocaval shunt [26]. In 1959, Bernstein and Cheiker developed the portosystemic shunt, which led portal blood into one of the iliac veins after functional hepatectomy [27]. In small animals, many other shunt techniques have been developed such as the portofemoral shunt or the mesentericocaval shunt via the jugular vein. In 1995, the splenocaval shunt was developed by Spiegel [28]. In large animals, on the other hand, a porto-femoro jugular bypass is frequently used [14, 29]. Results from experimental models of hepatic I/R injury alone are often extrapolated to clinical liver resection with PH and ischemia. However, in conventional experimental I/R models (for example, 70% partial hepatic ischemia), reperfusion ensues in the presence of nonischemic lobes [1]. Experimental models that combine PH and I/R injury rule out any contribution to recovery of the nonaffected liver tissue. Furthermore, in this model, postischemic recovery depends on the liver cell damage caused by the IR-injury and also on the stress caused by the liver resection and posthepatectomy liver regeneration [1, 30].

3.2.2 Global liver ischemia with spleen transposition

In 1970, Bengmark et al. developed this model for surgical treatment of portal hypertension [31]. In 1981, Meredith and Wade described a rat model that produced a portosystemic shunt in the anhepatic rat by transposition of the spleen, making a small incision in the left hypochondrium [32]. With the spleen inside a subcutaneous pouch, adequate portosystemic anastomoses emerge after some 2–3 weeks. The transposition induces the reversal of the blood flow in the splenic vein, which stimulates angiogenesis. Two weeks later, in the second step, a median laparotomy and temporary occlusion of the hepatoduodenal ligament are performed. This decompression by spleen transposition is easy to perform, because it does not require microsurgery. Within 2 or 3 weeks of surgery, the spleen is encapsulated without any signs of inflammation or bleeding. One drawback of this model is the long time span (3 weeks) until adequate portosystemic collaterals are large enough to take full control of portal vein flow. Furthermore, the effect of the changes in hepatic inflow on the collaterals remains unclear [6, 14, 33].

3.2.3 Partial hepatic resection under vascular occlusion

In 1982, Yamauchi et al. reported a hepatic ischemia model in which ischemia is induced by occlusion of the hepatic artery, the portal vein, and the bile duct of the left and median lobes. No extracorporeal shunt is required because the blood continues to flow through the right and caudal liver lobes. This model of partial ischemia (70%) has been extensively used in experimental studies of hepatic I/R [34–36]. An experimental model of 30% partial liver ischemia has also been used, in which occlusion at the hepatic artery and portal vein interrupts the supply of blood to the right lobe of the liver [19]. In the clinical setting, PH under I/R is normally performed to control bleeding during parenchymal dissection [6]. Therefore, an experimental model incorporating both hepatic regeneration and I/R injury can simulate the clinical situation of selective or hemihepatic vascular occlusion for liver resections. In this model, after left hepatic lobe resection, a microvascular clamp is placed across the portal triad supplying the median lobe (30%). Congestion of the bowel is prevented during the clamping because the portal flow through the right and caudate lobes is preserved. At the end of ischemia, the right lobe and caudate lobes are resected, and the clamp is released to achieve reperfusion of the median lobe. In this hepatic resection

model, portal decompression is not required, and certain important criteria are also met, such as reversibility, good reproducibility, and ease of execution [14, 19, 37].

3.3 Strategies applied in experimental models of normothermic ischemia

Many experimental studies have set out to develop in vivo pharmacological strategies for inhibiting the harmful effects of warm I/R [38–46]. Some of these studies are summarized in **Table 1**. However, none of them have been able to prevent hepatic I/R injury [6, 14]. However, it is important to develop strategies in experimental models that reproduce clinical practice conditions as closely as possible: for example, the use of intermittent clamping, and the combination of PH and I/R injury. Few of the studies carried out to date have complied with these requirements [12]. Some of these studies are summarized in **Table 2**. Recent breakthroughs in molecular biology are providing new opportunities for applying gene therapy to reduce liver I/R injury. The experimental data, however, have highlighted several problems inherent in gene therapy, including vector toxicity, difficulties in increasing transfection efficiency and protein expression at the appropriate site and time, and the difficulty of obtaining adequate mutants.

Warm ischemia		
Mice		
Drug	**Ischemic time**	**Effect**
Cerulenin	15 min	↓ UPC2, ↑ ATP
Platinum nanoparticles		↓ Hepatic injury
Exendin 4	20 min	↓ Hepatic injury and autophagy
Catalase and derivatives	30 min	↓ Oxidative stress
Apocynin		↓ Oxidative stress
Allopurinol		↓ Oxidative stress
N-Acetylcysteine	40, 90 min	↓ Hepatic injury, oxidative stress and apoptosis
Dipyridamole	45 min	↓ Hepatic injury
15-deoxy-$\Delta^{12,14}$-prostaglandin J_2	60 min	↓ Hepatic injury and inflammation
Ago-miR-46a		↓ Hepatic injury and apoptosis
Cold-inducible RNA-binding protein (CIRP) blockade		↓ Hepatic injury, apoptosis and inflammation
Anti-CD25 antibody		↓ Hepatic injury and inflammation
Diannexin		↓ Hepatic injury and inflammation
Ethyl pyruvate		↓ Hepatic injury, apoptosis and autophagy
Fasting		↓ Hepatic injury and inflammation; ↑ autophagy
Angiotensin II receptor antagonist		↓ Hepatic injury, apoptosis and inflammation
Riboflavin		↓ Hepatic injury, oxidative stress and inflammation
α7 Nicotinic acetylcholine receptor agonist		↓ Hepatic injury, oxidative stress and inflammation
Omega−3 Fatty acid		↓ Hepatic injury and inflammation; ↑ liver regeneration
Cobalt protoporphyrin	60, 90 min	↓ Hepatic injury and inflammation

Warm ischemia		
Mice		
Drug	**Ischemic time**	**Effect**
Hydroxytyrosol	75 min	↓ Hepatic injury, apoptosis, oxidative stress and inflammation
miR-370 inhibitor		↓ Hepatic injury and inflammation
Augmenter of liver regeneration (ALR)	90 min	↓ Hepatic injury, apoptosis and inflammation
Carbon monoxide		↓ Hepatic injury
Pan-selectin antagonist		↓ Inflammation
Erythropoietin		↓ Hepatic injury and apoptosis
Helium		↓ Hepatic injury; ↑ survival
Low-dose LPS		↓ Hepatic injury, apoptosis and inflammation
Protease-activated receptor 4 antagonist		↓ Hepatic injury, apoptosis and inflammation
Vasoactive intestinal peptide neuropeptide		↓ Hepatic injury, apoptosis and inflammation
Rats		
Drug	**Ischemic time**	**Effect**
ACE inhibitor	30 min	↓ Oxidative stress
ROS scavenger		↓ Apoptosis
Branched-chain amino acid (BCAA)		↓ Hepatic injury and inflammation
Carvacrol		↓ Hepatic injury, apoptosis and oxidative stress
CR2-CD59 (complement inhibitor)		↓ Hepatic injury; ↑ regeneration and survival
Hydrolysed whey peptide		↓ Hepatic injury, apoptosis and inflammation
Hyperbaric oxygen therapy		↓ Hepatic injury
Sivelestat sodium hydrate		↓ Hepatic injury and inflammation
Liraglutide		↓ Hepatic injury, apoptosis and inflammation
Allopurinol	30, 60 min	↓Oxidative stress
Diazoxide		↓ Hepatic injury and inflammation
PPARα agonist	30, 60, 90 min	↓ Oxidative stress and inflammation; ↑ autophagy
Propofol73		↓ Hepatic injury and apoptosis
Melatonin	35, 40 min	↓ Hepatic injury, apoptosis and oxidative stress
Levosimendan	40, 60 min	↓ Hepatic injury, apoptosis, oxidative stress and inflammation
Carnosic acid	45 min	↓ Hepatic injury
Limonin		↓ Hepatic injury, oxidative stress and inflammation
Low-intensity laser therapy		↓ Hepatic injury and oxidative stress
Rho-kinase inhibitor		↓ Hepatic injury; ↑ survival
Cardamonin		↓ Hepatic injury, oxidative stress and inflammation
Quercetin		↓ Hepatic Injury
Protoporphyrin		↓ Hepatic Injury

SOD	45, 60 min	↓ Inflammation
L-arginine		↓ Inflammation
Tocopherol	45, 90 min	↓ Oxidative stress and inflammation
IL-10	60 min	↓ Oxidative stress and inflammation
Anti-ICAM-1		↓ Inflammation
Gabexate mesilate		↓ Inflammation
Analogue of prostacyclin (OP-2507)		↓ Inflammation
n-3 PUFA		↓ Hepatic injury and oxidative stress
Adiponectin		↓ Hepatic injury, apoptosis and inflammation
Atorvastatin		↓ Hepatic injury and inflammation
Dexmedetomidine		↓ Hepatic injury and oxidative stress
Dioscin		↓ Hepatic injury, apoptosis and inflammation
Fibrin-derived peptide Bβ15–42		↓ Hepatic injury and inflammation
L-α-glycerylphosphorylcholine (GPC)		↓ Hepatic injury and oxidative stress
Rapamycin		↓ Hepatic injury; ↑ autophagy
Rosmarinic acid		↓ Hepatic injury, oxidative stress and inflammation
Sevoflurane		↓ Hepatic injury and oxidative stress
Simvastatin		↓ Hepatic injury, apoptosis and inflammation
Crocin		↓ Hepatic injury and oxidative stress
Tert-butylhydroquinone		↓ Inflammation
Hydrogen-rich saline		↓ Hepatic injury and oxidative stress
Spermine NONOate	60, 90 min	↓ Oxidative stress and inflammation
FK506		↓ Inflammation
Chloroquine		↓ Hepatic injury and inflammation
Lithium		↓ Hepatic injury and inflammation
AMPK activators	90 min	↑ NO, ATP
α-Lipoic acid		↓ Apoptosis; ↑ liver regeneration
Edaravone		↓ Hepatic injury and oxidative stress
Reduced glutathione		↓ Hepatic injury, apoptosis and oxidative stress
Sildenafil		↓ Hepatic injury, apoptosis and inflammation
Oleanolic		↓ Hepatic injury; ↑ survival
Unacylated-ghrelin		↓ Oxidative stress
Minocycline	2, 6 and 24 h	↓ Hepatic injury, oxidative stress and inflammation
Pigs		
Drug	**Ischemic time**	**Effect**
Sevoflurane	40 min	↓ Hepatic injury

Table 1.
Pharmacological strategies used in experimental models of warm ischemia.

Warm ischemia with partial hepatectomy		
Mice		
Drug	**Ischemic time**	**Effect**
Low dose of 2-complement receptor 1-related protein Y	30 min	↓ Hepatic injury and oxidative stress; ↑ liver regeneration
Melatonin	60 min	↓ Hepatic injury; ↑ liver regeneration
C1 esterase inhibitor		↓ Hepatic injury; ↑ liver regeneration and survival
Hydrogen sulfide	75, 90 min	↓ Hepatic injury and apoptosis; ↑ survival and liver regeneration
Rats		
Drug	**Ischemic time**	**Effect**
CF102	10 min	↓ Hepatic injury, apoptosis and inflammation; ↑ liver regeneration
Inchinkoto	15, 30 min	↓ Hepatic injury, oxidative stress and inflammation
Anti-HMGB1	20 min	↓ Hepatic injury; ↑ liver regeneration
Thrombomodulin		↓ Hepatic injury and apoptosis; ↑ liver regeneration
2mercaptoethane sulfonate	30 min	↓ Hepatic injury and oxidative stress; ↑ liver regeneration
Butyrate	30, 60 min	↓ Hepatic injury and inflammation
Omega3 fatty acids	40 min	↓ Hepatic injury and oxidative stress
Polyamines		↓ Necrosis, apoptosis and inflammation; ↑ liver regeneration
Glucose or lipid emulsion	60 min	↓ Hepatic injury; ↑ liver regeneration
Resistin or anti-visfatin antibodies		↓ Hepatic injury; ↑ liver regeneration
Tauroursodeoxycholate		↓ Oxidative stress
Sirolimus		↓ Inflammation
Combined angiotensin II receptor type 1 and 2 antagonists		↓ Hepatic injury and oxidative stress; ↑ liver regeneration
Thymoquinone		↓ Hepatic injury, apoptosis and oxidative stress
M3 AChR antagonist		↓ Hepatic injury and inflammation; ↑ regeneration and survival
Hydrogen sulfide	75, 90 min	↓ Hepatic injury, apoptosis and inflammation; ↑ regeneration and survival
IL-1 receptor antagonist	90 min	↓ Oxidative stress
Pigs		
Drug	**Ischemic time**	**Effect**
Hydrogen inhalation	20 min	↓ Hepatic injury, apoptosis and oxidative stress
Desferrioxamine	150 min	↓ Oxidative stress; ↑ liver function
Dogs		
Drug	**Ischemic time**	**Effect**
FK 3311 (Cox-2 inhibitor)	60 min	↓ Inflammation

Table 2.
Pharmacological strategies used in experimental models of warm ischemia with partial hepatectomy.

4. Controversies on hepatic regeneration under warm or cold ischemia

In an attempt to expand the size of the donor pool, the use of living related liver transplantation (LDLT) has helped increase the number of donor livers, but, nonetheless, concerns persist about graft-size disparity and hepatic regeneration. In 1990, Broelsch et al. reported the first 20 series of LDLT in the USA [47]. In 1997, Lo et al. [48] performed the first successful LDLT using an extended right lobe from a living donor for an adult recipient [6]. In LDLT, liver graft must be successfully regenerated; however, cold I/R, which will take place during liver transplantation surgery, will reduce the regenerative capacity of the liver.

The clinical application of strategies designed at beachside will depend on the use of experimental models that resemble as much as possible the clinical conditions in which the strategy intends to be applied [12]. However, many investigators have used rodent models of PH under or without vascular occlusion to mimic some of the pathophysiological events that occur during LT [6]. To the best of our knowledge, pharmacological strategies, which were used in experimental models of hepatic regeneration under warm ischemia (**Table 2**), were not applied in experimental models of LDLT. However, only three drugs (sirolimus, Ang II receptor type 2 antagonist, and Omega-3) were applied in patients submitted to LDLT (see **Table 3**). In contrast with the benefits on liver regeneration observed in experimental models of PH under I/R [49], the administration of sirolimus in LDLT decreases liver regeneration in patients [50]. Indeed, sirolimus decreases liver injury in patients only in combination with cyclosporine [51]. Similarly, angiotensin II receptor type 2 antagonist does not reduce hepatic injury as opposed to the benefits obtained in preclinical studies of PH under vascular occlusion [13, 52]. By contrast, pharmacological treatment with omega-3 had benefits on hepatic injury in clinical LDLT and in preclinical studies after PH under vascular occlusion [53, 54]. In our view, these controversial results may be explained at least partially, by the differences in the mechanism responsible of I/R damage and liver regenerative failure dependently on the surgical procedure (LDLT versus PH + I/R). Of note, it would be extremely useful to make a clear distinction between the mechanisms for each surgical situation to design therapies that demonstrate its effectiveness under experimental conditions similar to what happens in clinical practice [12]. This will probably lead to translation of the best strategies to clinical practice in the short term [12].

Living donor liver transplantation		
Human		
Drug	**Ischemic time**	**Effect**
Sirolimus	Not reported	↓ Liver regeneration
Sirolimus + cyclosporine		↓ Hepatic injury
Angiotensin II receptor type 2 antagonist		↑ Hepatic injury
Omega3		↓ Hepatic injury; ↑ liver regeneration

Table 3.
Pharmacological strategies used in living donor liver transplantation.

5. Conclusions

Although our knowledge about the mechanisms involved in the development of liver I/R injury and regenerative failure has significantly improved, and it has consequently been accompanied by a long list of potential therapeutic alternatives, I/R injury and regenerative failure after surgical procedure still represent a serious problem in the clinical practice. It should be considered that the mechanisms involved in hepatic I/R and regenerative failure very much depend on the experimental conditions used: which type of research is done, type of ischemia applied (warm or cold), period of ischemia (ranging from minutes to days), extension of hepatic ischemia (partial or total), graft subclinical situation (healthy, steatotic, aged,...), etc. Thus, new therapeutic strategies from experimental studies should be considered specific to the concrete experimental/surgical conditions used, and most probably, they cannot automatically be validated for all clinical situations requiring both vascular occlusion and liver regeneration [7]. We recognize the complication, but multidisciplinary research groups should devote additional efforts to better understand the cellular alterations and the crosstalk within the liver during the different clinical setting, requiring both vascular occlusion and liver regeneration to ultimately develop effectual therapeutic strategies aimed at reducing I/R damage and improving hepatic regeneration after liver surgery.

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Conflict of interest

The authors declare that they have no conflict of interest.

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References

[1] Casillas-Ramirez A, Escobedo-Medina SG, Cordero-Pérez P, Jiménez-Castro MB, Peralta C. Ischemia-reperfusion injury and oxidative stress. In: Gracia, Salvado, editors. Gastrointestinal Tissue: Oxidative Stress and Dietary Antioxidants. London: Elsevier INC; 2016. pp. 141-154

[2] Fu P, Li W. Nitric oxide in liver ischemia-reperfusion injury. In: Muriel P, editor. Liver Pathophysiology. London: Elsevier INC; 2017. pp. 125-127

[3] Clavien P, Harvey P, Strasberg S. Preservation and reperfusion injuries in liver allografts. An overview and synthesis of current studies. Transplantation. 1992;**53**:957-978

[4] Huguet C, Gavelli A, Chieco P, Bona S, Harb J, Joseph J, et al. Liver ischemia for hepatic resection: Where is the limit? Surgery. 1992;**111**:251-259

[5] Serafin A, Rosello-Catafau J, Prats N, Xaus C, Gelpi E, Peralta C. Ischemic preconditioning increases the tolerance of fatty liver to hepatic ischemia-reperfusion injury in the rat. The American Journal of Pathology. 2002;**161**:587-601. DOI: 10.1016/S0002-9440(10)64214-9

[6] Jiménez-Castro MB, Elias-Miró M, Casillas-Ramirez A, Peralta C. Experimental models in liver surgery. In: Abdeldayem, editor. Hepatic Surgery. Rijeka: InTech Open; 2013. pp. 121-166

[7] Peralta C, Jiménez-Castro MB, Gracia-Sancho J. Hepatic ischemia and reperfusion injury: Effects on the liver sinusoidal milieu. Journal of Hepatology. 2013;**59**:1094-1106. DOI: 10.1016/j.jhep.2013.06.017

[8] Selzner N, Rudiger H, Graf R, Clavien P. Protective strategies against ischemic injury of the liver. Gastroenterology. 2003;**125**:917-936

[9] Jaeschke H. Molecular mechanisms of hepatic ischemia-reperfusion injury and preconditioning. American Journal of Physiology. Gastrointestinal and Liver Physiology. 2003;**284**:G15-G26. DOI: 10.1152/ajpgi.00342.2002

[10] Montalvo-Jave EE, Escalante-Tattersfield T, Ortega-Salgado JA, Piña E, Geller DA. Factors in the pathophysiology of the liver ischemia-reperfusion injury. The Journal of Surgical Research. 2008;**147**:153-159. DOI: 10.1016/j.jss.2007.06.015

[11] Gracia-Sancho J, Villarreal G Jr, Zhang Y, Yu JX, Liu Y, Tullius SG, et al. Flow cessation triggers endothelial dysfunction during organ cold storage conditions: Strategies for pharmacologic intervention. Transplantation. 2010;**90**:142-149. DOI: 10.1097/TP.0b013e3181e228db

[12] Gracia-Sancho J, Casillas-Ramírez A, Peralta C. Molecular pathways in protecting the liver from ischaemia/reperfusion injury: A 2015 update. Clinical Science (London, England). 2015;**129**:345-362. DOI: 10.1042/CS20150223

[13] Ramalho FS, Alfany-Fernandez I, Casillas-Ramirez A, Massip-Salcedo M, Serafín A, Rimola A, Arroyo V, Rodés J, Roselló-Catafau J, Peralta C. Are angiotensin II receptor antagonists useful strategies in steatotic and nonsteatotic livers in conditions of partial hepatectomy under ischemia-reperfusion? The Journal of Pharmacology and Experimental Therapeutics. 2009;**329**:130-140. DOI: 10.1124/jpet.108.147835

[14] Mendes-Braz M, Elias-Miró M, Jiménez-Castro MB, Casillas-Ramírez A, Ramalho FS, Peralta C. The current state of knowledge of

hepatic ischemia-reperfusion injury based on its study in experimental models. Journal of Biomedicine & Biotechnology. 2012;**2012**:298657. DOI: 10.1155/2012/298657

[15] Katoonizadeh A. Liver Regeneration. In: Muriel P, editor. Liver Pathophysiology. London: Elsevier INC; 2017. pp. 113-123

[16] Mao SA, Glorioso JM, Nyberg SL. Liver regeneration. Translational Research. 2014;**163**:352-362. DOI: 10.1016/j.trsl.2014.01.005

[17] Ito K, Ozasa H, Horikawa S. Effects of prior splenectomy on remnant liver after partial hepatectomy with Pringle maneuver in rats. Liver International. 2005;**25**:438-444. DOI: 10.1111/j.1478-3231.2005.01102.x

[18] Gasbarrini A, Borle AB, Farghali H, Bender C, Francavilla A, Van Thiel D. Effect of anoxia on intracellular ATP, Na+i, Ca2+i, Mg2+i, and cytotoxicity in rat hepatocytes. The Journal of Biological Chemistry. 1992;**267**:6654-6663

[19] Peralta C, Bartrons R, Riera L, Manzano A, Xaus C, Gelpí E, Roselló-Catafau J. Hepatic preconditioning preserves energy metabolism during sustained ischemia. American Journal of Physiology. Gastrointestinal and Liver Physiology. 2000;**279**:G163-G171. DOI: 10.1152/ajpgi.2000.279.1.G163

[20] Caraceni P, Domenicali M, Vendemiale G, Grattagliano I, Pertosa A, Nardo B, et al. The reduced tolerance of rat fatty liver to ischemia reperfusion is associated with mitochondrial oxidative injury. The Journal of Surgical Research. 2005;**124**:160-168. DOI: 10.1016/j.jss.2004.10.007

[21] Cornide-Petronio ME, Casillas-Ramirez A, Jiménez-Castro MB, Peralta C. Experimental brain death models in liver transplantation. In: Tsoulfas, editor. Organ Donation and Transplantation. London: InTechOpen; 2018. pp. 135-151

[22] Yahanda A, Paidas C, Clemens M. Susceptibility of hepatic microcirculation to reperfusion injury: A comparison of adult and suckling rats. Journal of Pediatric Surgery. 1990;**25**:208-213

[23] Gasbarrini A, Addolorato G, Di Campli C, Simoncini M, Montemagn M, Castagneto M, et al. Gender affects reperfusion injury in rat liver. Digestive Diseases and Sciences. 2001;**46**:1305-1312

[24] Vidal M. Traitement chirurgical des ascites. La Presse Médicale. 1903;**11**:747-749

[25] Blakemore A, Lord J. The technic of using vitallium tubes in establishing portacaval shunts for portal hypertension. Annals of Surgery. 1945;**122**:449-475

[26] Burnett W, Rosemond G, Weston J, Tyson R. Studies of hepatic response to changes in blood supply. Surgical Forum. 1951;**94**:147-153

[27] Bernstein D, Cheiker S. Simple technique for Porto-caval shunt in the rat. Journal of Applied Physiology. 1959;**14**:467-470

[28] Spiegel H, Bremer C, Boin C, Langer M. Reduction of hepatic injury by indomethacin-mediated vasoconstriction: A rat model with temporary splenocaval shunt. Journal of Investigative Surgery. 1995;**8**:363-369

[29] Uhlmann D, Armann B, Gaebel G, Ludwig S, Hess J, Pietsch U, et al. Endothelin A receptor blockade reduces hepatic ischemia/reperfusion injury after warm ischemia in a pig model. Journal of Gastrointestinal Surgery. 2003;**7**:331-339

[30] He S, Atkinson C, Qiao F, Cianflone K, Chen X, Tomlinson S. A complement-dependent balance between hepatic ischemia/reperfusion injury and liver regeneration in mice. The Journal of Clinical Investigation. 2009;**119**:2304-2316. DOI: 10.1172/JCI38289

[31] Bengmark S, Börjesson B, Olin T, Sakuma S, Vosmic J. Subcutaneous transposition of the spleen: An experimental study in the rat. Scandinavian Journal of Gastroenterology. Supplement. 1970;**7**:175-179

[32] Meredith C, Wade D. A model of portal-systemic shunting in the rat. Clinical and Experimental Pharmacology & Physiology. 1981;**8**:651-652

[33] Suzuki S, Nakamura S, Sakaguchi T, Mitsuoka H, Tsuchiya Y, Kojima Y, et al. Pathophysiological appraisal of a rat model of total hepatic ischemia with an extracorporeal portosystemic shunt. The Journal of Surgical Research. 1998;**80**(1):22-27. DOI: 10.1006/jsre.1998.5419

[34] Hasselgren P, Jennische E, Fornander J, Hellman A. No beneficial affect of ATP-MgCl2 on impaired transmembrane potential and protein synthesis in liver ischemia. Acta Chirurgica Scandinavica. 1982;**148**:601-607

[35] Massip-Salcedo M, Zaouali M, Padrissa-Altés S, Casillas-Ramírez A, Rodés J, Roselló-Catafau J, Peralta C. Activation of peroxisome proliferator-activated receptor-alpha inhibits the injurious effects of adiponectin in rat steatotic liver undergoing ischemia-reperfusion. Hepatology. 2008;**47**:461-472. DOI: 10.1002/hep.21935

[36] Casillas-Ramírez A, Amine-Zaouali M, Massip-Salcedo M, Padrissa-Altés S, Bintanel-Morcillo M, Ramalho F, et al. Inhibition of angiotensin II action protects rat steatotic livers against ischemia-reperfusion injury. Critical Care Medicine. 2008;**36**:1256-1266. DOI: 10.1097/CCM.0b013e31816a023c

[37] Palmes D, Spiegel H. Animal models of liver regeneration. Biomaterials. 2004;**25**:1601-1611

[38] Elias-Miro M, Massip-Salcedo M, Jiménez-Castro MB, Peralta C. Does adiponectin benefit steatotic liver transplantation? Liver Transplantation. 2011;**17**:993-1004. DOI: 10.1002/lt.22358

[39] Fernández L, Heredia N, Grande L, Gómez G, Rimola A, Marco A, et al. Preconditioning protects liver and lung damage in rat liver transplantation: Role of xanthine/xanthine oxidase. Hepatology. 2002;**36**:562-572. DOI: 10.1053/jhep.2002.34616

[40] Serafin A, Rosello-Catafau J, Prats N, Gelpi E, Rodes J, Peralta C. Ischemic preconditioning affects interleukin release in fatty livers of rats undergoing ischemia/reperfusion. Hepatology. 2004;**39**:688-698. DOI: 10.1002/hep.20089

[41] Casillas-Ramírez A, Ben Mosbah I, Ramalho F, Rosello-Catafau J, Peralta C. Past and future approaches to ischemia-reperfusion lesion associated with liver transplantation. Life Sciences. 2006;**79**:1881-1894. DOI: 10.1016/j.lfs.2006.06.024

[42] Ben Mosbah I, Alfany-Fernández I, Martel C, Zaouali M, Bintanel-Morcillo M, Rimola A, et al. Endoplasmic reticulum stress inhibition protects steatotic and non-steatotic livers in partial hepatectomy under ischemia-reperfusion. Cell Death & Disease. 2010;**1**:e52. DOI: 10.1038/cddis.2010.29

[43] Bahde R, Spiegel H. Hepatic ischaemia-reperfusion injury from bench to bedside. The British Journal

of Surgery. 2010;**97**:1461-1475. DOI: 10.1002/bjs.7176

[44] Zúñiga J, Cancino M, Medina F, Varela P, Vargas R, Tapia G, et al. N-3 PUFA supplementation triggers PPAR-α activation and PPAR-α/NF-κB interaction: Anti-inflammatory implications in liver ischemia-reperfusion injury. PLoS One. 2011;**6**:e28502. DOI: 10.1371/journal.pone.0028502

[45] Ghonem N, Yoshida J, Stolz D, Humar A, Starzl T, Murase N, Venkataramanan R. Treprostinil, a prostacyclin analog, ameliorates ischemia-reperfusion injury in rat orthotopic liver transplantation. American Journal of Transplantation. 2011;**11**:2508-2516. DOI: 10.1111/j.1600-6143.2011.03568.x

[46] Kamo N, Shen X, Ke B, Busuttil R, Kupiec-Weglinski J. Sotrastaurin, a protein kinase C inhibitor, ameliorates ischemia and reperfusion injury in rat orthotopic liver transplantation. American Journal of Transplantation. 2011;**11**:2499-2507. DOI: 10.1111/j.1600-6143.2011.03700.x

[47] Broelsch C, Emond J, Whitington P, Thistlethwaite J, Baker A, Lichtor J. Application of reduced-size liver transplants as split grafts, auxiliary orthotopic grafts, and living related segmental transplants. Annals of Surgery. 1990;**212**:368-375

[48] Lo C, Fan S, Liu C, Lo R, Lau G, Wei W, et al. Extending the limit on the size of adult recipient in living donor liver transplantation using extended right lobe graft. Transplantation. 1997;**63**:1524-1528

[49] Arias-Diaz J, Ildefonso JA, Muñoz JJ, Zapata A, Jiménez E. Both tacrolimus and sirolimus decrease Th1/Th2 ratio, and increase regulatory T lymphocytes in the liver after ischemia/reperfusion. Laboratory Investigation. 2009;**89**:433-445. DOI: 10.1038/labinvest.2009.3

[50] Toso C, Patel S, Asthana S, Kawahara T, Girgis S, Kneteman NN, Shapiro AM, Bigam DL. The impact of sirolimus on hepatocyte proliferation after living donor liver transplantation. Clinical Transplantation. 2010;**24**:695-700. DOI: 10.1111/j.1399-0012.2009.01159.x

[51] Shinke H, Hashi S, Kinoshita R, Taniguchi R, Sugimoto M, Matsubara K, Ogawa E, Sonoda M, Takada N, Yoshizawa A, Ogawa K, Okamoto S, Uemoto S, Masuda S. Effectiveness of sirolimus in combination with cyclosporine against chronic rejection in a pediatric liver transplant patient. Biological & Pharmaceutical Bulletin. 2013;**36**:1221-1225

[52] Guillaud O, Gurram KC, Puglia M, Lilly L, Adeyi O, Renner EL, Selzner N. Angiotensin blockade does not affect fibrosis progression in recurrent hepatitis C after liver transplantation. Transplantation Proceedings. 2013;**45**:2331-2336. DOI: 10.1016/j.transproceed.2013.01.067

[53] Marsman HA, Heger M, Kloek JJ, Nienhuis SL, ten Kate FJ, van Gulik TM. Omega-3 fatty acids reduce hepatic steatosis and consequently attenuate ischemia-reperfusion injury following partial hepatectomy in rats. Digestive and Liver Disease. 2011;**43**:984-990. DOI: 10.1016/j.dld.2011.07.009

[54] Ibrahim ES, Saleh SM, El Hoseeny M, El Shaarawy A. Effect of omega-3 on hepatic regeneration in adult living donors undergoing hepatic resections for liver transplantation: A randomized controlled trial. Journal of Critical Care. 2016;**31**:157-162. DOI: 10.1016/j.jcrc.2015.09.022.