A Study on Liver Intercellular Adhesion Molecule-1 (ICAM-1) Induction Including Sepsis, Warm Ischaemia, Cold Ischaemia And Reperfusion Injury

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Abstract: Introduction: Intercellular adhesion molecule-1 (ICAM-1), a member of the immunoglobulin-gene-superfamily, is constitutively expressed on vascular endothelium and mediates leukocyte-endothelial cell interaction. Lipopolysaccharides (LPS) or endotoxin activates complement and signalling mechanisms in both macrophages and endothelial cells to produce inflammatory mediators. These mechanisms stimulate ICAM-1 induction. Ischaemia reperfusion inury induces the same reaction like LPS. Objective: To investigate ICAM-1 expression on sinusoidal endothelial cells (SEC's) in normal rat liver after induction of sepsis, ischaemia (warm and cold), Ischaemia/reperfusion injury on both ischaemic and non ischaemic liver lobes. Methodology: Male Sprague Dawley (SD) rats weighing 200-250g were used in the experimental studies. All experiments were non recovery. We designed three experimental models. Induction of sepsis model, in situ warm ischaemia reperfusion, and cold ischaemia models. Results & conclusion: Treated animals with LPS had intense expression of ICAM-1 in both SEC's and hepatocytes. Warm ischaemia over 45min produced marked ICAM-1 expression on SEC's, this expression bacame more intense after a different period of reperfusion. However, cold ischaemia up to 8hrs has no significant effect of ICAM-1 expression. Reperfusion of the post cold ischemic liver resulted in significant upregulation of ICAM-1 expression. In conclusion, according to the results in this study which showed the up regulation of ICAM-1 expression as a secondary to the inflammatory process which strats immediately after the ischaemic injury and increased after ischaemia/reperfusion injury.

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1. Introduction

Intercellular adhesion molecule-1 (ICAM-1), a member of the immunoglobulin-gene-superfamily, is constitutively expressed on vascular endothelium and mediates leukocyte-endothelial cell interaction by binding to the beta-integrins CD11a/CD18 (LFA-1) and CD11b/CD18 (Mac-1) [1,2].

Inflammatory reaction results in a rapid upregulation in ICAM-1 expression on endothelial cells.[3] The central event in the inflammation reaction is the activation of circulating leukocytes which must first adhere to endothelial cells, then migrate between adjacent endothelial cells and finally move through the extra cellular matrix to perform effector functions (leukocyte-endothelial cell interaction.[4,5]

Ischaemia and reperfusion of the liver is associated with transient hepatic dysfunction, presumably due to the high susceptibility of the liver to ischaemia and cell injury additionally induced by reperfusion.[6] The mechanism involved in the development of ischaemia/reperfusion hepatic disorders have to be considered as complex in nature based on the direct interaction of intravascular blood cells, i.e. leukocytes and platelets, with non parenchymal cells, i.e. endothelial and Kupffer cells, and indirect communication between the different cellular compartments, both releasing and responding to a variety of potent mediators, such as cytokines and oxygen drived-free radicals.[7-10] The understanding of this complex interaction is of major interest not only to extend knowledge on the pathophysiology of hepatic ischaemia/reperfusion injury but also to establish novel therapeutic approaches counteracting the manifestation of tissue injury.[8]

Lipopolysaccharides (LPS) or endotoxin has been shown to be responsible for the pathophysiology associated with gram-negative sepsis.[11-13] LPS activates complement and signalling mechanisms in both macrophages and endothelial cells to produce inflammatory mediators.[14,15] The hepatic macrophages, the Kupffer cells (Kc) release a large number of these inflammatory mediators and are considered the major source of circulating mediators e.g. tumour necrosis factor (TNF-α), interleukins (IL-1, IL-6 and IL-8) and reactive free radicals or their precursors.[16] All of these substances may directly and/or indirectly affect hepatic parenchymal and sinusoidal endothelial cells (SEC's) as well as circulating blood elements resulting in an inflammatory response and tissue injury. The initial manifestation of this injury is characterised by

activation of the endothelium with increased adhesiveness for leukocytes and platelets.[11,16]

Oxidative stress is defined as an increase in the production of reactive oxygen species which has been related to ischaemia/reperfusion injury.[17] It was shown that reperfusion of the ischaemic isolated rat liver produces sufficient quantities of reactive oxygen species to produce structural cell damage.[18] This event called a vascular oxidant stress, has been identified as being generated by the resident macrophages of the liver (Kc).[19] Reactive oxygen species generated during reperfusion has been implicated by numerous pharmacological intervention studies to be involved in hepatic reperfusion injury.[18,20] Reduced glutathione (GSH) is one of the components of the UW solution and it is important in cellular defence against oxidative stress.[21] Induction of ischaemia of rat liver for 90 min resulted in decrease of 18.3% of reduced glutathione. Subsequent reperfusion for 60 min resulted in marked decrease in reduced glutathione.[22,23] Hypothermic preservation of rat hepatocyte in modified UW solution (no GSH) for 24hrs resulted in more than 50% loss of intracellular GSH. The addition of GSH (3 mmol/L) to the UW solution improves the viability of hepatocytes stored more than 48hrs, as was assessed by measuring ATP content and cellular membrane integrity.[24]

The Objectives of this study are: To investigate ICAM-1 expression on normal rat liver cells (SEC's and hepatocytes) and after induction of sepsis. Invetigate the effect of ischaemia (warm and cold) and Ischaemia/reperfusion injury on ICAM-1 expression on both ischaemic and non ischaemic liver lobes.

2.Methodology

Animals

Male Sprague Dawley (SD) rats weighing 200-250g were used in the experimental studies. The animals had free access to a standard diet and tap water prior to the experiment. All experiments were non recovery and were carried out at Experimental Unit, Theodor Bilharz Reasearch Institute from October 2010 – June 2011. Tissue biopsies were analysed and processed for ICAM-1 assessment as previously reported. [1-5]

Experimental groups

a) Induction of sepsis

Group1 : normal control
Group2 : saline group
Group3: treated group (injection of LPS intraperitoneal)

b) In situ warm ischaemia and ischaemia/reperfusion

Group1 :sham operation for two hours
Group2 : 30min. ischaemia
Group3 : 45min. ischaemia.
Group4 : 60min. ischaemia.
Group5 : 90min. ischaemia.
Group6 : 105min. ischaemia.
Group7 : 120min. ischaemia.
Group8 : 30min. ischaemia followed by 60min. reperfusion.
Group9 : 45min. ischaemia followed by 60min. reperfusion.
Group 10: 60min. ischaemia followed by 60min. reperfusion.

c) Cold ischaemia and cold ischaemia followed by warm reperfusion.

Group 1 : 0hr cold storage time
Group 2 : 8hr cold ischaemia
Group 3 : 16hr cold ischaemia
Group 4 : 24hr cold ischaemia
Group 5 : 0hr cold ischaemia/60min reperfusion.
Group 6 : 8hr cold ischaemia/60min reperfusion.
Group 7 : 16hr cold ischaemia/60min reperfusion.
Group 8 : 24hr cold ischaemia/60min reperfusion.

Surgical procedures

Three different non-recovery animal models were used in this study. Wedge liver biopsies were taken in all models. Laparotomy was carried out via a midline ventral abdominal skin incision. Blood supply (portal and arterial) to the left and median hepatic lobes were identified.

Animal body temperature (BT), heart rate beat/min (HR, bpm) and tissue oxygen saturation (O_2 sat.) were continuously monitored during the experimental procedure.

a) Induction of septic shock model

Experiment conducted on 18 Sprague Dawley (SD) male rats (200-250 g) in three groups of 6 animals each were used in this model. In the first group (normal control group) animals underwent laparotomy under general anaesthesia and liver biopsies were taken. In the second group 5 ml/kg body weight of normal saline was injected intraperitoneally. In the third group 5 ml/kg body weight of lipopolysaccharides (LPS) was administered intraperitoneally to induce sepsis like effect.[11] Saline and LPS were injected without anaesthesia and there was no mortality in either groups after the injection. In the saline and the treated groups liver biopsies were taken 3 hours after the administration of either LPS or saline.

b) *In situ* warm ischaemia and ischaemia/reperfusion (I/R)

In situ warm ischaemia was induced by complete occlusion of the blood supply (arterial and portal) to the left and median hepatic lobes using a vascular clamp for different time periods. In the I/R groups subsequent reperfusion was obtained by releasing the clamp for 60 minutes. Liver biopsies were taken immediately following laparotomy (baseline biopsy),

post-ischaemia and post-reperfusion from the ischaemic lobe (left lobe). Additional post-reperfusion biopsies were taken from the non-ischaemia lobe (right lobe) to investigate the systemic effect of reperfusion injury on ICAM-1 expression in this lobe.

c) Isolated perfused rat liver (IPRL)

Livers were retrieved from rats. After hepatectomy livers were stored in cold UW solution for different periods (0, 8, 16 and 24 hours). At the end of cold storage time, livers were perfused in non-recirculating technique. Liver biopsies were taken on laparotomy (baseline), after cold storage, and at the end of reperfusion time.

Investigation of ICAM-1 expression

In all models liver biopsies were snap frozen in liquid nitrogen and processed for ICAM-1 expression. ICAM-1 expression on sinusoidal endothelial cells (SEC's) and hepatocytes was assessed by light microscopy without the knowledge of the treated groups. The staining intensity on sinusoidal endothelial cells (SEC's) and hepatocytes was scored semiquantitatively as reported in previous studies.[15,25-29] According to stain intensity ICAM-1 expression was graded into four categories, no or faint stain of ICAM-1, mild, moderate and intense ICAM-1 staining, these grades were scored from 0-3 respectively.[30] Plates 1, 2, 3 and 4.

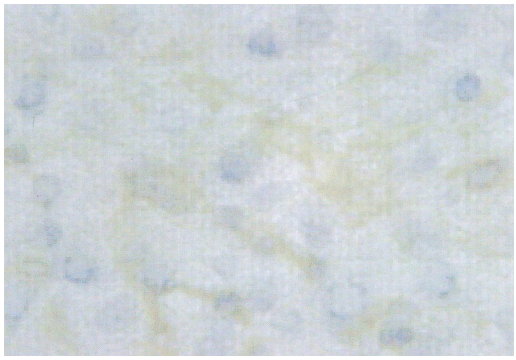

Plate 1. Faint ICAM-1 staining (light brown pigmentation) on SEC's in normal rat liver (X100).

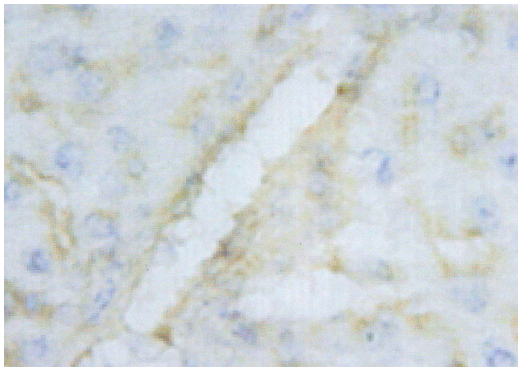

Plate 2. Mild ICAM-1 staining (brown pigmentation) on SEC's (X100).

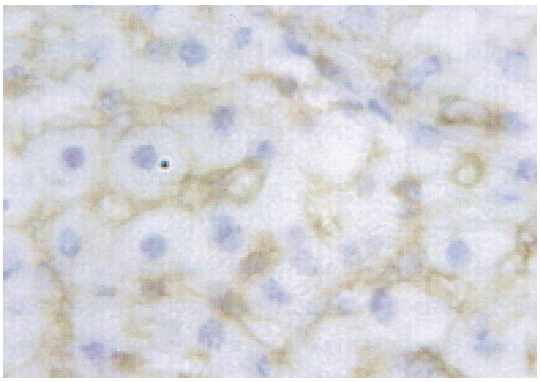

Plate 3. Moderate ICAM-1 staining (brown pigmentation) on SEC's, no staining was observed on hepatocytes (X100).

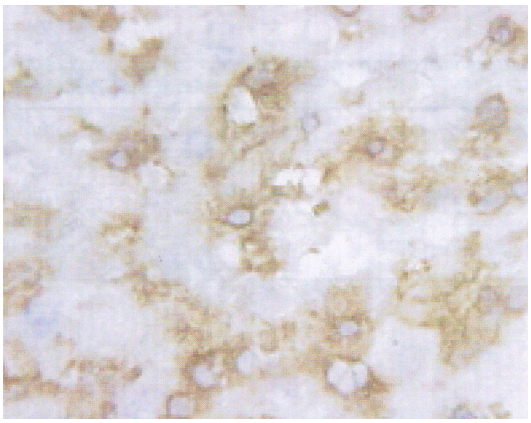

Plate 4. Intense ICAM-1 staining (brown pigmentation) on SEC's and hepatocytes (X100).

3. Results

In all experiments there was no animal death during the experimental protocol. There were no differences between groups in HR, BT and O_2 sat. during the conduction of the experiments.

Effect of intraperitoneal injection of LPS toxin on ICAM-1 expression

The results are summarised in table 1. Sections from normal controls (group 1) showed that 67% of the animals had no ICAM-1 expression. In 33% there was mild ICAM-1 expression on sinusoidal endothelial cells (SEC's), but no ICAM-1 expression on hepatocytes was observed.

ICAM-1 expression was similar in the saline treated group with 50% animals showing no ICAM-1 expression, and the other 50% mild expression on SEC's. Again there was no hepatocyte expression of ICAM-1.

In group 3 (LPS group) all treated animals (100%) had an intense ICAM-1 expression on the SEC's and hepatocytes. ICAM-1 expression had a score of 3.

There was a significant up regulation of ICAM-1 expression in group 3 when compared with group1 and group 2, p<0.0001 (one way analysis of variances).

Table 1. ICAM-1 expression in normal rat liver and after intraperitoneal injection of saline and LPS. Results are expressed as Mean±SE.

Groups	ICAM-1 expression
Group1 (normal control)	0.33±0.21
Group2 (saline group)	0.5±0.22
Group3 (LPS group)	2.83±0.17

Effect of warm ischaemia on ICAM-1 expression.

In between groups there were no differences on ICAM-1 expression in baseline liver biopsies.

Warm ischaemia for 30 min did not induce ICAM-1 expression on SEC's. However warm ischaemia for 45 min or more did produce sufficient induction of ICAM-1 on SEC's.

In these ischaemic groups ICAM-1 expression was limited on the SEC's, no expression was observed on the hepatocytes.

There was also significant difference in ICAM-1 expression in the post ischaemia groups 3,4, 5, 6 and 7 when compared with the expression in the sham operated group, $p<0.05$ (one way analysis of variances). Results summarised in Table 2.

Table 2. Baseline ICAM-1 expression and following warm ischaemia. Results are expressed as Mean±SE and *P* value.

Warm ischaemia period		Base line	Following warm ischaemia	*p* value
Sham operated	(group1)	0.17±0.17	0.50±0.22	ns (not significant)
30min	(2)	0.17±0.17	0.50±0.34	ns
45min	(3)	0.33±0.21	1.00±0.29	<0.05
60min	(4)	0.33±0.21	1.33±0.21	<0.05
90min	(5)	0.17±0.17	1.50±0.34	<0.05
105min	(6)	0.17±0.17	1.50±0.22	<0.05
120min	(7)	0.17±0.17	1.67±0.33	<0.05

Effect of warm ischaemia/reperfusion on ICAM-1 expression

a) Effect of ischaemia/reperfusion injury on the ischaemic lobe (left hepatic lobe)

With 30min of warm ischaemia followed by 60min reperfusion (group8), 45min I/R and 60min I/R (groups 9 & 10) there was a significant increase in ICAM-1 expression on SEC's when compared with baseline biopsies in each group, $p<0.05$.

In these groups ICAM-1 expression was moderate-intense on SEC's and hepatocytes.

There was a significant up regulation in SEC's ICAM-1 expression in the post reperfusion groups when compared with the sham operated group, $p<0.0001$ (one way analysis of variances). The results are summarised in table 3.

There was also significant difference in ICAM-1 expression after ischaemia/reperfusion when compared with the expression after warm ischaemia only.

Table 3. ICAM-1 expression at baseline and post-reperfusion liver biopsies. Results are expressed as Mean±SE and P value.

Ischaemia/reperfusion period	Laparotomy	Post reperfusion	*P* value
Sham operation	0.17±0.17	0.50±0.22	ns
Group 8 (30min isch./6omin rep.)	0.17±0.17	1.30±0.21	<0.05
Group 9(45min isch./60min rep.)	0.33±0.21	2.50±0.22	<0.05
Group 10(60min isch./60min rep.)	0.17±0.17	2.80±0.17	<0.05

b) Effect of I/R on the non ischaemic lobe (right hepatic lobe)

ICAM-1 expression in the non-ischaemic lobe (right lobe) in group 8 which had 30min ischaemia followed by 60min reperfusion was analysed. The result was compared with ICAM-1 expression in the sham operated group and there was no significant difference between the two groups.

In group 9 (45min ischaemia/60min reperfusion) and group 10 (60min ischaemia/60min reperfusion) there was significant ICAM-1 expression up- regulation in the non ischaemic lobe when compared with the expression in the sham operated group($p<0.05$).

Effect of cold ischaemia on ICAM-1 expression

There was no difference in ICAM-1 expression on SEC's in livers who had 0 and 8 hours cold ischaemia times. There was also no difference noted when compared with the controls. With increasing the cold ischaemia time to 16 hours and above there was up regulation of ICAM-1 expression on the SEC's when compared with the normal control group.

There was a statistical difference in ICAM-1 expression when the overall result for the cold ischaemia groups was compared with the normal control group, $p < 0.05$ (one way analysis of variances). Results are summarised in table 4.

Table 4. Effect of cold ischaemia on ICAM-1 expression. Results are expressed as Mean±SE and P value.

Cold ischaemia time	Post isch.	Normal control (Mean±SE)	*P* value
0 h (in situ liver flush)	0.67 ± 0.33	0.33±0.21	ns
8 h cold ischaemic time	0.83 ± 0.31	0.33±0.21	ns
16 h cold ischaemic time	1.17 ± 0.31	0.33±0.21	<0.05
24 h cold ischaemic time	1.83 ± 0.31	0.33±0.21	<0.05

Effect of on bench reperfusion on ICAM-1 expression

There was an up regulation of ICAM-1 expression after cold ischaemia/warm reperfusion when compared with the expression at the beginning of laparotomy (baseline expression) in each group.

With the increase of cold ischaemia time to 16 hours or more and followed by 60min warm reperfusion, the ICAM-1 expression became more significant when compared with groups with cold ischaemia less than 16 hours.

There was also a highly significant increase in ICAM-1 in the post reperfusion biopsy when compared with the expression in the control group, $p<0.0001$ (one way analysis of variances). Results are summarized in table 5.

Table 5. ICAM-1 expression at baseline biopsies and after ischaemia/reperfusion. Results are expressed as Mean±SE and *P* value.

Cold ischaemia/reperfusion time	Baseline ICAM-1 expression	Postreperfusion ICAM-1 expression.	*P* value
Group 5 (in situ liver flush./60min rep.)	0.17±0.17	1.50±0.34	<0.05
Group 6 (8h cold isch./60min rep.)	0.33±0.21	2.17±0.31	<0.05
Group 7 (16h cold isch./60min rep.)	0.17±0.17	2.67±0.21	<0.0001
Group 8 (24h cold isch./60min rep.)	0.17±0.17	2.98±0.02	<0.0001

4.Discussion

In this study three different animal models were used to study interaction, features for liver ICAM-1 expression.

In the first model LPS endotoxin was injected into the intraperitoneal cavity to induce a sepsis like effect. The second model is the regional ischaemia/reperfusion model in which blood flow to the left and median lobes of the liver was occluded using a vascular clamp whereas flow to the right lobes remained intact. Reperfusion was obtained by removing the vascular clamp. The third model was the isolated perfused rat liver model. ICAM-1 is known to be modulated by various inflammatory mediators such as interleukin-1 (IL-1), tumour necrotic factor-α (TNF-α), interferon-γ and increased endotoxin level.[31-35]

LPS is mainly cleared from the portal blood by the liver (endothelial cells and macrophages) and causes the same clinical features as can be observed in sepsis.[36] Vollmar and colleagues reported accumulation of leukocytes, interacting with the endothelial lining of both sinusoids and post sinusoidal venules in rat liver exposed to LPS.[14]

In this study ICAM-1 showed a weak staining or no staining of the SEC's in normal rat liver and no evidence of hepatocytes ICAM-1 expression. This result confirms previous reports.[29,37,38] In another in vitro study it was shown that there was weak staining of ICAM-1 on the hepatocytes.[39]

In this study the treated animals with LPS had intense expression of ICAM-1 in both SEC's and hepatocytes 3 hours after LPS administration. On the hepatocytes there was diffuse membranous and focal cytoplasmic expression of ICAM-1. This up regulation of ICAM-1 expression was observed previously on the SEC and on the hepatocytes as well using same LPS dose and same animal model.[11] Significant up regulation of ICAM-1 expression in the different rat organs has been reported after IP administration of S. abortus equi to produce an endotoxemia that has the same pathophysiology as LPS endotoxemia.[38] ICAM-1

expression up regulation was reported 3-5 hours after endotoxin administration.

This dramatic up regulation of ICAM-1 expression could be explained by a direct effect of LPS on the Kc and SEC's. LPS activates Kc and SEC's which participate in the inflammatory response by releasing reactive oxygen intermediates and a variety of cytokines (IL-1, IL-8 and TNF-α). TNF-α levels increase significantly 1 hour after LPS exposure and is followed by PMNs infiltration of the sinusoids which peaks after the plasma TNF-α level.[37,40]

The pathophysiologic mechanism of cellular injury that follows ischaemia and reperfusion is still incompletely understood. I/R of the liver is associated with transient hepatic dysfunction of the liver, presumably due to the high susceptibility of the liver to ischaemic cell injury additionally induced by reperfusion.[41-43] The development of the post-ischaemic hepatic inflammatory response is complex in nature, involving leukocytic activation by potent chemotactic mediators, cell adhesion to the endothelial lining via distinct adhesion molecules and the action of additional mediators released by the adherent leukocytes.[44,45] Previous experimental studies have demonstrated that leukocytes adhere to endothelial cells of both sinusoids and postsinusoidal venules after warm as well as cold ischaemia and ischaemia/reperfusion.[10,46,47] ICAM-1 has been shown to be responsible to mediate adherence between leukocytes and the endothelial cells.[46,48]

In this study 30 or 45min of warm ischaemia did not induce ICAM-1 expression but 60-120min produced marked SEC's ICAM-1 expression. The hepatocytes exhibited no ICAM-1 expression. The mechanism of ICAM-1 expression during warm ischaemia is mainly due to Kc activation.[49-51] Activated Kc produce various cytokines that lead to infiltration of polymorphonuclear leukocytes (PMNs) into the post-ischemic liver.[29] SEC's is the first target of cytotoxic products released from Kc, this is followed by transendothelial migration and neutrophil accumulation at the site of inflammation which requires Mac-1 (CD11b/CD)-ICAM-1 interaction.[29,52,53] This followed by up regulation of ICAM-1 expression on the SEC's.

The duration of warm ischaemia affects survival. In a study of hepatic ischaemia in rats, the extend of warm ischaemia from 30min to 60min affect the survival outcome, as all recipients of allografts with 30 min of warm ischaemia lived more than 22 days and all recipient with 60 min warm ischaemia died within 2 days.[50]

The central role of ICAM-1 induction in warm ischaemia injury to the liver has been demonstrated by the survival improvement after the administration of anti-ICAM-1 antibody after the onset of the ischaemia.[52] In the present study it was shown that warm ischaemia up to 45min did not induce significant ICAM-1 expression.

Cold storage of transplanted organs reduces metabolic pathways and delays pathological changes.[54] Ultrastructure studies conducted on cold ischaemic rat liver (after 2, 4, 6, 10, 16, 24 and 48 hours cold ischaemia) have demonstrated that the first preservation damage was in the SEC's which became rounded and swollen after 16 hr of cold ischaemia. Loss of viability of SEC's as determined by morphological criteria was complete after 24 hrs or longer and Kc structure was altered dramatically. However, after 24 hr there was no significant change in the hepatocytes structure.[55-57]

In the present study up to 8 hrs cold ischaemia had no influence on ICAM-1 expression on SEC's. With cold ischaemia of 16-24 hrs there was significant induction of ICAM-1 expression. Previous report has shown no evident of ICAM-1 expression in rat livers after 1 and 6 hr cold storage.[58] ICAM-1 expression in the cold preserved liver in the current study confirms the previous published data which shown that 0 hr preserved liver released a negligible amount of TNF-α whereas 24 hr preserved liver produced the highest amount of TNF-α.[59]

Reperfusion of the post-ischemic liver resulted in definable and quantifiable changes in leukocyte kinetics. It has been shown that the number of infiltrating PMNs is significantly increased by short periods of ischaemia (20min) being followed by reperfusion for 2, 12 and 24 hours.[60] These changes are likely to be due to upregulation of endothelial cell adhesion molecules.[60] It was also previously reported that I/R (60min ischaemia followed by 60min reperfusion) induced rolling of leukocytes to the endothelial lining of postsinusoidal venules. In addition, leukocytes were found stagnant in hepatic sinusoids.[61]

In the current study warm ischaemia for 30, 45 and 60min followed by 60min reperfusion resulted in a highly significant ICAM-1 expression on both SEC's and hepatocytes when compared with the control. This ICAM-1 expression was intense on SEC's and hepatocytes. There was also significant difference in ICAM-1 expression after ischaemia/reperfusion when compared with the expression after warm ischaemia only.

Reperfusion following cold ischaemia has been shown to activate Kc resulting in elevated levels of IL-1 and TNF-α.[62,63] In the current study cold ischaemia up to 8 hr followed by warm reperfusion for 60min induced intense ICAM-1 expression on both SEC's and hepatocytes. With extended cold ischaemia time up to 16 and 24 hr as would be found with the liver transplantation, this produced a highly significant up regulation of ICAM-1 expression on the SEC's and hepatocytes. These results are in agreement with the previously published data which showed an early induction of ICAM-1 expression on SEC's 15min post-reperfusion in livers who subjected to 6hr cold.[64]

In another experimental animal model, isolated rat livers were stored for 8 or 24 hrs and this was followed by 15 min of reperfusion.[56] Nuclear trypan blue uptake and lactate dehydrogenase release were used as indices of hepatocyte and SEC's viability. Loss of hepatocyte viability was nearly undetectable. SEC's loss of viability averaged about 4% after 8 hrs and 30 % after 24hrs of storage and brief reperfusion.[56] This observation fits with the results in the current study In livers stored for 16 hrs and 24 hrs and followed by 60min warm reperfusion the ICAM-1 expression increased more than in 0 hr and 8 hrs cold ischaemia followed by 60min reperfusion. So, longer periods of cold storage (16 + 24hrs) are associated with SEC's ICAM-1 induction and are known from previous studies to be associated with loss of endothelial cell viability.[56]

In the present study ICAM-1 expression was up-regulated in the non-ischaemic lobe after 60min ischaemia followed by 60min reperfusion. This ICAM-1 expression was intense on the SEC's and weak on the hepatocyte. The ICAM-1 expression in the non-ischaemic lobe would suggest that activate Kc and SEC's during I/R produce a general inflammatory response which in turn is responsible for the ICAM-1 induction of the non-ischaemic liver lobe.[52]

According to the results in this study which showed the up regulation of ICAM-1 expression as a secondary to the inflammatory process which strats immediately after the ischaemic injury and increased after ischaemia/reperfusion injury. This phenomenon might be very important in the field of liver transplantation as it could be used as an early signe of the inflammatory cascad that startes in the transplanted liver graft. Accordingly it would allow to start a new regim of immunosuppresion to prevent the rejection at the cellular level of the transplanted liver graft. This needs more stduies using anti ICAM-1 for further evaluation of its immunological impact.

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