Functional recovery after liver resection
Veteläinen, R.L.

Citation for published version (APA):

General rights
It is not permitted to download or to forward/distribute the text or part of it without the consent of the author(s) and/or copyright holder(s), other than for strictly personal, individual use, unless the work is under an open content license (like Creative Commons).

Disclaimer/Complaints regulations
If you believe that digital publication of certain material infringes any of your rights or (privacy) interests, please let the Library know, stating your reasons. In case of a legitimate complaint, the Library will make the material inaccessible and/or remove it from the website. Please Ask the Library: http://uba.uva.nl/en/contact, or a letter to: Library of the University of Amsterdam, Secretariat, Singel 425, 1012 WP Amsterdam, The Netherlands. You will be contacted as soon as possible.
Summary and conclusions
Summary and conclusions

In chapter 1, an outline of the thesis is presented and a brief overview of aspects of liver steatosis in general is given.

In chapter 2, the features are reviewed in relation to the increased susceptibility to injury in liver surgery. There are numerous conditions connected with induction of hepatic steatosis. After the initial hepatocyte fat accumulation, a complex intracellular cascade is activated. This cascade includes production and accumulation of reactive oxygen species causing structural damage to several intracellular organelles, most importantly mitochondria. Additionally, activation of hepatic Kupffer cells induces a vicious circle of progressive inflammation and hepatocellular damage. These pathogenic features have special implications for the postoperative recovery of patients after hepatic resection and contribute to increased ischemia-reperfusion injury and affected hepatocyte proliferation. These contributors play, according to current knowledge, crucial roles in the increased susceptibility of steatotic livers to injury.

The exact clinical significance of the type and extent of steatosis remains unclear as larger cohort studies applying uniform diagnostic criteria and histopathological assessment are missing. The gold standard of diagnosis is still histopathological evaluation of several biopsies as none of the state of the art radiological modalities is specific enough to detect all histopathological features of steatosis needed for staging and evaluation of prognosis. Further studies, using uniform diagnostic criteria and larger cohorts, are of great need in the future.

In chapter 3, we show the outcome of two different non-alcoholic fatty liver disease (NAFLD) models induced by choline deficient and methione-choline deficient diets, respectively, in a rat model. The choline deficient (CD) diet fed rats developed mainly microvesicular steatosis and only occasionally inflammatory cells were present in the liver parenchyma. Furthermore, the rats developed features closely resembling the human metabolic syndrome, i.e. obesity, dyslipidemia and insulin resistance. In contrast, in the methione-choline deficient (MCD) diet fed rats plasma lipid levels were undetectable, insulin homeostasis was unaffected and body weight decreased. Steatosis in MCD-fed rats was mainly of the macrovesicular type and progression to steatohepatitis was observed at the end of the experimental period of 7 weeks. This progression was possibly attributable to the increased lipid peroxidation and decreased antioxidant levels compared to no changes in the CD-fed rats. As a conclusion from chapter 3, it can be stated that although equivalence for the outcome of both MCD and CD diets can be found in the clinical situation, the results of models applying these diets should be compared with caution.

In chapter 4, we describe in detail the utility of the nuclear imaging techniques, 99mTc-GSA scintigraphy and 99mTc-mebrofenin HBS in liver surgery including liver resections and transplantation. In contrast to conventional techniques for the assessment of liver function, i.e. liver volume by computed tomography, these applications give information on both total as regional liver function. This analysis enables a reliable and more accurate
definition of the safety limits of liver resection and a selection of the high-risk patients as conventional liver volume assessment does not always correlate with function. Two nuclear imaging techniques can be potentially applied in the estimation of the severity of parenchymal liver disease, in the follow-up of liver regeneration after portal vein embolization or in the noninvasive follow-up of liver function patients with transplanted liver grafts. Both 99mTc-GSA scintigraphy and 99mTc-mebrofenin HBS are directly applicable in liver surgery and liver transplantation. However, the major disadvantages of 99mTc-GSA scintigraphy are the poor availability outside Japan and the complex data analysis method. However, the advantage is that 99mTc-GSA scintigraphy is minimally affected by changes in hepatic circulation compared to other blood clearance tests. 99mTc-mebrofenin HBS is readily available and the analysis is simple and highly reproducible. The disadvantages are the potential bias from high plasma bilirubin concentrations and changes in hepatic circulation.

In chapter 5, the excellent reproducibility of calculations of hepatobiliary function by dedicated animal pinhole hepatobiliary scintigraphy (HBS) using 99mTc-mebrofenin (r = 0.95, P < 0.001) is demonstrated. The dynamic acquisition of scintigraphy was adapted to the faster metabolism in rats. All calculations for 99mTc-mebrofenin uptake rate were made during equal blood distribution of 99mTc-mebrofenin, between 30 and 120 sec after injection of the radiopharmaceutical, which is before the average Tpeak of maximal hepatic 99mTc-mebrofenin. HBS correlated well with liver mass at baseline and after 70% partial hepatectomy (PH) (r = 0.94, P < 0.001 and r = 0.85, P < 0.001, respectively). There was a good, but less strong association between liver weight and Tpeak or T½ peak after PH (r = 0.78, P < 0.01 or r = 0.73, P < 0.05, respectively). This might be due to more difficult data analysis of Tpeak and T½ peak. These results support the utility of HBS in non-invasive imaging of hepatocellular function, providing both visual and quantitative information. Furthermore, serial measurements can be performed in one animal enabling a longitudinal study design which obviously decreases inter-subject variation and the amount of animals needed.

In chapter 6, the utility of 99mTc-GSA with SPECT is shown for the assessment of liver function and volume in a rat model. The hepatic binding of 99mTc-GSA was homogenous in all liver lobes and the data analysis was highly reproducible (r = 0.783, P = 0.017). In normal rat livers, there was a strong and significant correlation between functional volume assessed by 99mTc-GSA SPECT and conventional liver volume (r = 0.93 P < 0.0001). In regenerating liver after 70% PH, the mean liver functional volume and conventional liver volume decreased 1 day compared to baseline and regenerated to baseline level at day 5. There was a strong correlation between the two volumes in the regenerating liver (r = 0.86, P < 0.0001). One day after 70% PH, the 99mTc-GSA uptake significantly decreased compared to baseline and linearly increased in 7 days to baseline level However, when 99mTc-GSA uptake per liver weight (g) was analyzed, first at 1 day it decreased (vs baseline, P < 0.009) but remained at the same level 3 and 5 days after 70% PH and returned to baseline level on day 7. The results from chapter 6 show that 99mTc-GSA scintigraphy combined with SPECT is a feasible, non-invasive method to assess hepatic
volume in a normal and a regenerating rat liver. However, the hepatic 99mTc-GSA uptake underestimates the hepatic regeneration and might therefore not be accurate for the assessment of liver function in a regenerating rat liver.

In chapter 7, a significant correlation is shown between the severity of steatosis and parameters of hepatobiliary scintigraphy. The hepatic uptake rate of 99mTc-mebrofenin decreased and the time of maximal hepatic uptake of 99mTc-mebrofenin increased linearly while the severity of steatosis increased. There was a significant correlation between 99mTc-mebrofenin uptake calculations and biochemical and histopathological markers used for steatosis evaluation. There was no correlation of HBS with plasma markers of hepatocellular injury (transaminases) indicating that HBS calculations reflect actual hepatobiliary function rather than damage. These results suggest a potential role of 99mTc-mebrofenin scintigraphy as a noninvasive, functional follow-up method for development or regression of liver steatosis in patients.

In chapter 8, we show that the recovery of hepatocellular volume after partial hepatectomy (PH) is impaired in rats with severe steatosis. After PH, Kupffer cell-mediated inflammatory responses, both local and systemic, and hepatocellular injury were prolonged and increased in rats with severe steatosis. In contrast, this phenomenon was only transient in rats with mild steatosis and in control rats. Necrosis was the main type of cell death in rats with severe steatosis reflecting impaired restorative mechanisms after hepatocellular injury, i.e. after PH. Apoptosis, as seen in controls and rats with mild steatosis, is considered to be the physiological cell death pathway as only minimal inflammatory response is induced, in contrast to severe inflammation to necrosis, in the surrounding liver parenchyma. The impairment of liver regeneration was most likely due to aggravated hepatic lipid peroxidation and defective hepatocellular recovery mechanisms, mediated by IL-10 and antioxidant scavenging of reactive oxygen species. The results of this chapter suggest an increased risk of performing extensive liver resection in the presence of severe steatosis.

In chapter 9 we show impaired functional recovery of mild steatotic livers after liver resection. The recovery of actual liver volume after 70% liver resection, by hepatocyte proliferation, was similar in mild steatotic rats and controls as there was no difference in the increase in regenerating liver mass or hepatocyte proliferation index. However, hepatocellular damage, as measured by plasma transaminases, was aggravated in mild steatotic rats after resection. Also the functional recovery, as evaluated by HBS, was impaired in mild steatotic rats up to 7 days postoperatively. This prolonged functional recovery was probably attributable to mitochondrial dysfunction as in mild steatotic rats the recovery of hepatic energy balance, evaluated by hepatic adenosine triphosphate (ATP) levels, was delayed. Even though there was no difference in preoperative ATP levels between the groups, it is possible that due to underlying pathologic changes caused by fat accumulation, subclinical mitochondrial changes are present already preoperatively. The outcome of this study sheds light on the mechanisms of impaired recovery of mild steatotic livers after resection.
In chapter 10, it is demonstrated that portal vein ligation is as effective as sequential dual ligation of both hepatic artery and portal vein in inducing hepatocyte proliferation as only a transient increase in hepatocyte proliferation was seen at 24h when applying the latter. Both sequential and simultaneous dual ligation significantly increased the systemic proinflammatory response and local response in the regenerating liver compared to portal vein ligation. After portal vein ligation, the ligated liver parenchyma recovered after initial necrotic changes, in contrast to fibro-necrosis seen after 14 days of sequential dual ligation and complete necrosis after simultaneous dual ligation. These results suggest that for induction of liver regeneration portal vein ligation only is sufficient, however, if tumor destruction and/or decrease in tumor size is the primary goal, sequential dual ligation of both hepatic artery and portal vein might be useful.

In chapter 11, we investigated the possible association of a benign parenchymal liver disease, i.e. liver adenomatosis (LA), and steatosis. Using a Medline search between 1963 and 2006, we could identify 94 previously published patients with LA that fitted with the criteria defined by Flejou et al. 18% of all LA patients had steatosis in the non-tumoral part of the liver. The clinical importance of steatosis has been acknowledged only in recent years. Also, in recent years, the incidence of steatosis is increasing because of its association with Western lifestyle. To investigate this aspect we performed a search in our own patient database at the AMC. Interestingly, we could identify 6 patients with histologically confirmed LA. Of these six patients, four patients presented with NAFLD and one with NASH in the non-tumoral liver. It has been suggested that there is connection between liver steatosis and LA. Recently, a connection between deranged glucose metabolism, commonly seen in steatotic patients, and LA has been made via hepatocyte nuclear factor 1α. The management of LA patients remains a problem because of the rarity of the disease. Our review of literature, however, shows that primary clinical presentation can predict the presence of, intraperitoneal and intratumoral bleeding that can cause complications. Surgical management, either resection or even transplantation should only be applied in cases of aggressive tumor growth, or serious discomfort or hemorrhagic complications. Conservative management with careful patient follow-up is warranted for patients with less aggressive clinical presentation.