Microvascular Research 94 (2014) 47–51

Contents lists available at ScienceDirect

Microvascular Research

journal homepage: www.elsevier.com/locate/ymvre

Microcirculation changes during liver resection — A clinical study

Jan Nilsson [a,1], Sam Eriksson [a,1], Per-Jonas Blind [a], Pehr Rissler [b], Christian Sturesson [a,*]

[a] Department of Surgery, Clinical Sciences Lund, Lund University and Skåne University Hospital, Lund, Sweden
[b] Department of Pathology, Clinical Sciences Lund, Lund University and Skåne University Hospital, Lund, Sweden

ARTICLE INFO

Article history:
Accepted 5 May 2014
Available online 16 May 2014

Keywords:
Chemotherapy
Hepatic microcirculation
Liver function
Sidestream dark-field imaging
Steatosis

ABSTRACT

Background: In this study we aimed to evaluate effects of liver resection on hepatic microcirculation. In addition we wanted to study if histological liver damage could be detected intra-operatively.
Patients and methods: 40 patients undergoing hepatic resection were included and grouped according to if they were operated with a major or minor resection. Hepatic microcirculation measurements were made intra-operatively before and after liver resection with sidestream dark-field (SDF) imaging. Red blood cell velocity (RBCV), sinusoidal diameter and functional sinusoidal density were determined.
Results: After hepatic resection RBCV increased in both the minor and major groups (44 μm/s, P = 0.016 and 121 μm/s, P = 0.002). RBCV in patients with histological damages was 225 (148–464) μm/s vs. 161 (118–329) μm/s in patients with no damage (P = 0.016).
Conclusion: A hepatic resection leads to an increase of sinusoidal RBCV. SDF imaging could potentially be used to intraoperatively identify histological damages.

Introduction

Liver resection is currently the standard of care for liver tumor disease, with future liver remnant volume and function determining resectability (Adams et al., 2013; Poon et al., 2002). Currently large resections leaving as little as 20–25% of healthy liver are considered safe (Kishi et al., 2009). In larger resections there is a risk of postoperative liver insufficiency which is the single most important factor for postoperative mortality (Rahbari et al., 2011). However, pathological changes to the liver parenchyma, as liver cirrhosis or chemotherapy-induced damages, demand larger remnants to secure postoperative liver function.

Intraoperative assessment of hepatic microcirculation in liver transplants is reported predictive of early graft postoperative function (Puhl et al., 2005). Moreover, hepatic microcirculation is affected by liver parenchymal changes associated with chemotherapy for colorectal cancer in experimental animals (DeLeve et al., 2003; McCuskey et al., 2004; Seifalian et al., 1999). Similar studies on hepatic microcirculation in human liver resections have not been made. Preoperative chemotherapy can induce liver steatosis, steatohepatitis and sinusoidal obstruction syndrome (SOS), which have been shown to increase postoperative complications and worsen the long-term prognosis after a liver resection (Gomez et al., 2007; Peppercorn et al., 1998; Rubbia-Brandt et al., 2006; Tamandl et al., 2011; Vauthey et al., 2006). We hypothesize that human hepatic microcirculation is affected by liver parenchymal damages as well as a major liver resection.

Methods for experimental measurement of hepatic microcirculation traditionally include intravital fluorescence microscopy and laser-Doppler flowmetry (LDF) (Vollmar and Menger, 2009). Intravital fluorescence microscopy requires toxic fluorescent dyes for contrast enhancement making it impossible for use in a clinical setting. LDF is a non-invasive technique allowing real-time assessment of perfusion, but because of its high variable baseline signal and inter-site variability it can only measure relative blood perfusion (Vollmar and Menger, 2009). Another interesting method to assess hepatic microcirculation that we recently tested is laser speckle contrast imaging (LSCI), permitting fast non-contact measurements of a large surface area (Eriksson et al., accepted for publication; Sturesson et al., 2013).

In recent years the introduction of orthogonal polarization spectral (OPS) imaging (Groner et al., 1999) and its successor sidestream dark-field (SDF) imaging has been developed for direct visualization of microcirculation. SDF imaging consists of a hand-held microscope that can be directly positioned onto the tissue to visualize its microcirculation (Goedhart et al., 2007). Tissue illumination is accomplished by green light with a central wavelength of 530 nm, which is absorbed by hemoglobin-containing red blood cells (RBC) and reflected by surrounding tissue. SDF imaging renders sharper images than OPS imaging due to less blurring (Goedhart et al., 2007). Direct visualization allows measurement of sinusoidal RBC velocity (RBCV), sinusoidal diameter (SD) and functional sinusoidal density (FSD) (Langer et al., 2001; Puhl

Abbreviations: FSD, functional sinusoidal density; LDF, laser-Doppler flowmetry; LSCI, laser speckle contrast imaging; OPS, orthogonal polarization spectral; RBCV, red blood cell velocity; ROI, region of interest; SD, sinusoidal diameter; SDF, sidestream dark-field; SOS, sinusoidal obstruction syndrome.

* Corresponding author at: Department of Surgery, Skåne University Hospital, S-221 85 Lund, Sweden. Tel.: +46 46 172347; Fax: +46 46 172335.
E-mail address: christian.sturesson@med.lu.se (C. Sturesson).

[1] Contributed equally to the study.

http://dx.doi.org/10.1016/j.mvr.2014.05.002
0026-2862/

et al., 2003). Using SDF imaging, hepatic microcirculation has successfully been measured on rat liver (Sturesson et al., 2013).

In this study we aimed to evaluate effects of liver resection on hepatic microcirculation. In addition we wanted to study if SDF imaging can detect histological damages of the liver parenchyma.

Patients and methods

The study protocol was approved by the local ethics committee. All patients scheduled for liver resection between January 22, 2013 and June 4, 2013 at Skåne University Hospital, Lund, Sweden were considered for inclusion except patients with viral hepatitis and patients undergoing laparoscopic surgery. All included patients were informed about the study and gave their consent both orally and in writing. A total of 40 patients were included and they were grouped according to if they were operated with a major (n = 12) or minor (n = 28) resection. Major resection was defined as a right or extended right hepatectomy. Due to the small number of included patients, we had no intention to investigate mortality or morbidity.

Anesthesia and surgery

Anesthesia was induced by propofol and fentanyl and maintained with desflurane, isoflurane or sevoflurane, with or without nitrous oxide and fentanyl. No patient had epidural anesthesia. Positive end-expiratory pressure (PEEP) was initially set to 5 mm Hg. According to the surgeon's preference, the PEEP was set to zero during liver transection. The aimed central venous pressure (CVP) was 5 mm Hg or lower during liver parenchymal transection.

The liver became accessible through a right-sided subcostal incision with upward midline extension. Standard technique was used to perform hepatectomy (Blind et al., accepted for publication) and parenchymal transection was made using an ultrasound aspiration dissector (CUSA®, Valleylab Inc., Boulder, CO, USA). If necessary, a sling was placed around the hepatoduodenal ligament allowing the blood inflow vessels, consisting of the portal vein and hepatic artery, to be occluded by temporarily tightening the sling (Pringle's maneuver). After resection, the excised liver was immediately fixed in 4% formalin for histological analysis (see below).

SDF imaging

Three investigators (JN, SE, CS) performed all SDF imaging interventions. For intraoperative measurements, the tip of the SDF imaging microscope (MicroScan Video Microscope System, MicroScan BV, Amsterdam, The Netherlands) was covered with a sterile, 10 mm diameter, disposable lens cap (MicroScan Lens, MicroVision Medical, Amsterdam, The Netherlands) and the rest of the probe, including approximately 2 m of the cable system, was encased into sterile foil (Video camera laser drape, Microtek Medical B.V., Zutphen, The Netherlands).

Hepatic microcirculation measurements were incorporated in the standard liver resection surgery procedure and were performed twice per patient: first after the liver had been exposed and mobilized from its diaphragmatic attachments and second directly after liver resection had been made. Every measurement consisted of measuring on three locations (region of interest, ROI) (Boerma et al., 2005) on a liver area not to be resected, typically the center of Couinaud's segment three or five. Prior to measurements, the liver capsule was removed from an area of approximately two times 2 cm with the intention of getting the probe closer to the liver parenchyma and getting sharper pictures. If necessary room temperature sterile 0.9% NaCl solution was flushed gently on the liver parenchyma in order to remove coagulated blood and the surface was dried gently with a sterile dry cloth. Operating room lightning was switched off before recording began and the SDF imaging probe was gently applied manually to minimize pressure on the liver parenchyma and allowing stable pictures (Tytgat et al., 2013). Each ROI was recorded for 20 s during apnea. By using an analog–digital capture device (ADVC110, Grass Valley USA, LLC, San Francisco, USA), the image from the SDF imaging microscope was converted to a digital signal and then recorded (25 frames per sec) directly to a standard laptop hard drive using a video capture and vascular analysis software package (AVA 3.0, MicroScan BV, Amsterdam, The Netherlands). At the time of each measurement CVP, mean arterial blood pressure (MAP) and PEEP were noted.

Microcirculatory analysis

All microcirculatory analyses were performed by two investigators (JN, SE) using the same software package used for video capture (AVA 3.0). Before analyzing the video sequences, images were stabilized and quality enhanced by adjusting contrast and background gray level with the analysis software. Analysis of RBCV was made using automatically generated space–time diagrams (Dobbe et al., 2008) after manually identifying three randomly allocated vessels in each recorded sequence. Three vector lines were drawn in each diagram. After automatic vessel detection, determination of both mean SD (μm) and FSD (mm/mm^2) were made, the latter being calculated as length of perfused vessels per observation unit area. A mean for each patient was made and the following variables were defined: PreRBCV, PostRBCV (RBCV before and after resection), PreSD, PostSD (SD before and after resection) and PreFSD, PostFSD (FSD before and after resection). The differences between before and after resection values were calculated for each patient.

Histological analysis

Histological analyses were made using hematoxylin and eosin stain and trichrome stain by one and the same pathologist, who had no knowledge of the patient's clinical data. Steatosis was classified according to D'Alessandro et al. (1991) and steatosis was defined as grade ≥2. Steatohepatitis was graded according to Kleiner et al. (2005) using the Nonalcoholic fatty liver disease Activity Score, NAS. A NAS ≥4 was considered steatohepatitis. SOS was defined as a sinusoidal dilatation grade ≥2 according to Rubbia-Brandt et al. (2004). Fibrosis was graded according to Kleiner et al. (2005). A fibrosis grade ≥2 was considered significant fibrosis. Liver parenchyma damage was defined as any of steatosis, steatohepatitis, SOS or significant fibrosis.

Statistical analysis

All results are expressed as median (range). Correlations were made using linear regression analysis and computing a Pearson correlation coefficient, r for parametric variables and Spearman correlation coefficient, r_s for non-parametric variables. Mann–Whitney *U*-test was used to compare continuous data and Fisher's exact test was used for categorical data. Tests on related samples were performed using the Wilcoxon signed-rank test. A P-value of <0.05 was considered statistically significant. Statistical analysis was performed using IBM SPSS Statistics version 21 (IBM, Armonk, NY).

Results

Hepatic microcirculation measurements were successfully made in all 40 patients, resulting in analyzable SDF imaging film sequences in which flowing RBC could be clearly visualized in the sinusoids. Patient characteristics for the major and minor resection groups are shown in Table 1 and results regarding sinusoidal blood flow velocity, sinusoidal diameter and functional sinusoidal density are shown in Table 2. The total duration of the measuring procedure was about 5 min per patient and the time needed for subsequent analysis was about 30 min per patient. In Table 3, SDF imaging results are shown for patients with liver