Aspects of retransfusion of shed blood in cardiac surgery

Brondén, Björn

2011

Avdelningen för Anestesiologi och Intensivvård, Institutionen för Kliniska Vetenskaper i Lund
Medicinska Fakulteten, Lunds Universitet

Aspects of Retransfusion of Shed Blood in Cardiac Surgery

Akademisk avhandling

som med vederbörligt tillstånd från Medicinska Fakulteten vid Lunds Universitet för avläggande av doktorsexamen i medicinsk vetenskap i ämnet anestesiologi och intensivvård kommer att offentligen försvaras i Belfragesalen BMC (D1539a), Lund, fredagen den 20 maj 2011, kl. 13:00

av

Björn Brondén
Leg Läkare

LUNDS UNIVERSITET
Medicinska fakulteten

Handledare
Henrik Bjursten, Docent i Thoraxkirurgi, Lunds Universitet

Biträdande handledare
Sten Blomquist, Docent i Anestesi och Intensivvård, Lunds Universitet
Magnus Denker, Docent i Klinisk Fysiologi, Lunds Universitet

Fakultetsopponent
Anders Jeppsson, Professor i Thoraxkirurgi, Sahlgrenska Akademin, Göteborgs Universitet

Ordförande
Per-Olof Grände, Professor i Anestesi och Intensivvård, Lunds Universitet

Organization	Document name
LUND UNIVERSITY Faculty of Medicine Institution of Clinical Sciences in Lund Department of Anaesthesia and Intensive Care	DOCTORAL DISSERTATION
	Date of issue 2011-05-20
	Sponsoring organization

Author(s)
Björn Brondén

Title and subtitle

Aspects of retransfusion of shed blood in cardiac surgery

Abstract

The use of cardiotomy suction during open heart surgery with cardiopulmonary bypass (CPB) has a long tradition and often regarded as mandatory. Recently, studies have described potentially negative effects of cardiotomy suction with activation of the complement system, enhanced inflammatory response, hemolysis and coagulopathy. Emboli of various matters has also been studied. Whether they affect the function of various organs is an ongoing debate.

Renal dysfunction after cardiac surgery is a well known complication. Monitoring of renal function is important, both for the guidance of renal intervention and for the prediction of outcome. Cystatin C has been suggested as a more sensitive marker of glomerular filtration rate (GFR) than creatinine, which today is the most commonly used marker of GFR.

In study I and II, we studied the distribution and the kinetics of lipid microemboli during cardiac surgery in a porcine model. The organ that received most emboli was the kidney. Within the brain, the grey matter received most emboli. A high degree of first-pass trapping in the capillaries was found. No immediate renal excretion of lipid material was seen.

In study III, we evaluated cystatin C with iohexol clearance in a clinical study on patients scheduled for coronary artery bypass grafting (CABG). We found that cystatin C is suitable for monitoring renal function in cardiac surgery.

In study IV, the final part of this thesis, we launched a clinical trial including 150 patients, where we compared outcome between CABG with and without cardiotomy suction. There were a few differences, all favoring surgery without the use of cardiotomy suction. No difference was found regarding renal function. We could conclude that CABG with CPB can be conducted safely without using cardiotomy suction and retransfusion of shed blood as long as the surgeon is vigilant on the blood loss, and prepared to use cardiotomy suction when needed.

Key words: CABG, cardiotomy suction, lipid microemboli, porcine study, clinical study, cystatin C, creatinine, iohexol clearance, outcome, retransfusion, transfusion, blood.

Classification system and/or index termes (if any):

Supplementary bibliographical information:	Language English
ISSN and key title: 1652-8220 Aspects of retransfusion of shed blood in cardiac surgery	ISBN 978-91-86671-97-6

Recipient's notes	Number of pages	Price
	Security classification	

Aspects of retransfusion of shed blood in cardiac surgery

Björn Brondén, M.D.

Lund University
Faculty of Medicine

Department of Anaesthesia and Intensive Care

Sweden 2011

ISSN 1652-8220
ISBN 978-91-86671-97-6
Lund University, Faculty of Medicine Doctoral Dissertation Series 2011:49
Printed by Media-Tryck, Lund, Sweden 2011

Till Frida och Karolina

Contents

Summary 9

Sammanfattning (populärvetenskaplig) 11

Original Papers 13
- *Study I* 13
- *Study II* 13
- *Study III* 13
- *Study IV* 13

Abbreviations 15

Introduction 17
- Cardiotomy suction 18
- Liquid scintillation process 18
- Iohexol 19
- Cystatin C 19
- Creatinine 20

Aims of the Studies 21
- *Study I* 21
- *Study II* 21
- *Study III* 21
- *Study IV* 21

Material and Methods 23
- Study protocol 23
  - *Study I* 23
  - *Study II* 23
  - *Study III* 24
  - *Study IV* 24
- Anaesthesia in laboratory study (study I, II) 25
- Anaesthesia in clinical study (study III, IV) 25
- Administration of radioactive shed blood (study I, II) 26
- Processing of samples in laboratory study (study I, II) 26
- Iohexol clearance protocol (study III) 27
- Calculations 27

Statistical analysis 28
Study I 28
Study II 28
Study III 29
Study IV 29
Results 31
Study I 31
Study II 33
Study III 36
Study IV 40
Discussion 55
Conclusions 63
Study I 63
Study II 63
Study III 63
Study IV 63
Grants 65
Acknowledgements 67
References 69

Summary

This thesis is based on four studies. In a novel porcine model, we found that lipid material retransfused into the cardiopulmonary bypass (CPB) circuit during heart surgery reached the systemic circulation rapidly. This lipid material seemed to be trapped in all organs of the body to varying extent. The blood flow to an organ did not seem to be the sole factor determining the embolic load in that organ. In the brain, the organ of focus in research on lipid microembolization, we found that the grey matter of the cerebrum was the part of the brain that contained most lipid microemboli (LME). Since the gray matter of the cerebrum is involved in the cognitive functions of the brain, these findings will further spur the ongoing debate of the possible relationship between LME and postoperative cognitive dysfunction in cardiac surgery. The organ that contained most lipid material was the kidney. In daily clinical practice, it is common to encounter patients suffering from renal dysfunction after cardiac surgery. Therefore we became interested in the possibility of a relation between LME and postoperative renal dysfunction. Creatinine is the most commonly used marker for monitoring renal function. However, it has a rather low sensitivity, and is therefore not suitable to detect smaller changes in renal function. In this aspect, cystatin C is known to be a better biomarker. As a part of this thesis, we therefore evaluated plasma cystatin C with iohexol clearance for use in cardiac surgery, and found it suitable for monitoring renal function in cardiac surgery. In the final part of this thesis, we launched a clinical trial, where we compared outcome between coronary artery bypass grafting (CABG) with (which is routine practice) and without cardiotomy suction (i.e with and without retransfusion of shed mediastinal lipid containing blood). The study revealed some interesting findings. More transfusion of blood products were given to the control group that received retransfusion of shed blood, compared with the study group that did not receive shed blood. A few other minor differences and trends were found, all favoring surgery without the use of cardiotomy suction. The study did not find any differences between the groups regarding renal function. To summarize, we could conclude that CABG with CPB can be conducted safely without using cardiotomy suction and retransfusion of shed blood, as long as the surgeon is vigilant on the blood loss and prepared to use cardiotomy suction when needed.

Sammanfattning
(populärvetenskaplig)

De flesta större hjärtoperationer idag kräver att hjärtat under en del av operationen står stilla utan att blod cirkulerar igenom det. För att kunna bibehålla ett blodflöde av syresatt blod genom kroppen kopplas en hjärt-lungmaskin till patienten. Blodet förs via slangar från höger förmak först ner till ett uppsamlingskärl, varifrån blodet pumpas vidare genom en oxygenator i vilken blodet syresätts. Därefter pumpas blodet tillbaks in i kroppen via en slang kopplad till kroppspulsådern. På så vis upprätthålls cirkulationen av syresatt blod genom kroppen utan att hjärtat eller lungorna deltar. För att kunna göra detta krävs att blodet inte levrar sig, vilket åstadkoms genom att patienten gives stora mängder heparin under tiden som hjärt-lungmaskinen används. Operationsområdet är format som en skål, varför det blod som läcker ut under operationen blir kvar där. Eftersom detta sårblod inte levrar sig går det att återanvända. Ett sätt att göra detta är att suga upp sårblodet och föra det till uppsamlingskärlet i hjärt-lungmaskinen och därefter skicka tillbaks det in i blodcirkulationen. Återförande av sårblod på detta sätt förekommer idag vid de flesta hjärtoperationer som genomförs.

Genom tidigare studier vet man att det inte är helt komplikationsfritt att återanvända sårblod. Bland annat kan olika substanser såsom fett från benmärg och gasbubblor skickas ut i cirkulationen. I de arbeten som denna avhandling bygger på, ville vi bland annat studera vad som händer med det fett som skickas ut i blodcirkulationen. Vi fann att en stor del av det fett som skickades tillbaks ut i cirkulationen hamnade i njurarna (delarbete I och II). Man vet sedan tidigare att njurfunktionen ofta försämras efter en hjärtoperation. Vanligtvis återställs den, men inte alltid. Kunde fett som fastnar i njurarna (fettembolier) ha något med saken att göra? Vi behövde ett bra instrument för att i vardagskliniken kunna mäta njurfunktion på patienter som genomgår hjärtkirurgi. Vi fann att laboratorieprovet cystatin C var ett bra sådant instrument (delarbete III). Slutligen genomförde vi en klinisk studie med 150 deltagande patienter som alla genomgick bypassoperation (delarbete IV). Hälften av patienterna opererades med återanvändning av sårblodet och hälften opererades utan att sårblodet återanvändes. Vi ville veta om utfallet för patienterna efter operationen skilde

sig mellan grupperna. De flesta parametrar som undersöktes visade inte på några skillnader mellan grupperna. De skillnader som vi fann talade huvudsakligen till fördel för operation utan återanvändning av sårblod. Bland annat visade det sig att gruppen, i vilken sårblod återanvändes, behövde mer transfusion av blodprodukter under och efter operationen jämfört med den andra gruppen. Vi kunde efter studien fastslå att genomförande av bypassoperation utan återanvändning av sårblod kan genomföras säkert, om man bara är observant på blodförlusterna och återanvänder sårblodet om större blödning tillstöter. Vi fann inga skillnader mellan grupperna vad det gällde postoperativ njurfunktion.

Original Papers

This thesis is based on the following studies, which will be referred to in the text by their Roman numbers.

Study I
Bronden B, Dencker M, Allers M, Plaza I, Jonsson H. Differential distribution of lipid microemboli after cardiac surgery. Ann Thorac Surg 2006;81:643-8.

Study II
Bronden B, Dencker M, Blomquist S, Plaza I, Allers M, Jonsson H. The kinetics of lipid micro-emboli during cardiac surgery studied in a porcine model. Scand Cardiovasc J 2008;42:411-6.

Study III
Bronden B, Eyjolfsson A, Blomquist S, Dardashti A, Ederoth P, Bjursten H. Evaluation of cystatin C with iohexol clearance in cardiac surgery. Acta Anaesthesiol Scand 2011;55:196-202.

Study IV
Bronden B, Eyjolfsson A, Blomquist S, Dardashti A, Ederoth P, Bjursten H. Cardiac surgery with and without cardiotomy suction. A prospective observational study. Manuscript submitted for publication.

Abbreviations

ACT	activated clotting time
ALAT	alanine-aminotranspherase
ASAT	aspartate-aminotranspherase
AUC	area under curve
BSA	body surface area
CABG	coronary artery bypass grafting
CCS	Canadian Cardiovascular Society class
CG	Cockcroft-Gault
CPB	cardiopulmonary bypass
CKMB	creatine kinase subunit MB
COPD	chronic obstructive pulmonary disease
CRP	C-reactive protein
Da	dalton
DMP	disintegrations per minute
eCyC	estimated cystatin C clearance
GFR	glomerular filtration rate
LME	lipid microemboli
LVEF	left ventricular ejection fraction
IQR	inter quartile range
mCi	milliCurie
MDRD	the modification of diet in renal disease
NYHA	New York Heart Association class
SCADS	small capillary arteriolar dilatations
SD	standard deviation
X-over	cross-over

Introduction

The use of cardiotomy suction during open heart surgery with cardiopulmonary bypass (CPB) has a long tradition and is often regarded as mandatory [1]. It helps the surgeon to visualize the surgical field during larger bleedings without excessive blood loss, and cardiotomy suction can also be life-saving in difficult situations.

Recently, studies have described potentially negative effects of cardiotomy suction, where the focus has been on the activation of the complement system and enhanced inflammatory response [2-4]. In addition, embolic material of various matters such as lipids, air and bone fragments have been studied [3, 5-12]. Hemolysis [2, 3, 13] and coagulopathy have also been described [3, 14-16]. These conditions in the blood can potentially have effects on various organs [3, 17]. Neurological and renal dysfunctions after cardiac surgery are well known complications [18-21]. Whether these complications are related to the use of cardiotomy suction is an ongoing debate [17, 22].

Renal dysfunction after cardiac surgery is a common complication, with 1–5% of the patients requiring dialysis [18, 23]. It causes discomfort for the patients and generates an additional cost for the healthcare system. Moreover, a decline in renal function in conjunction with cardiac surgery affects long-term survival [24-26], as well as in other major surgery [27]. Monitoring of renal function is therefore important, both for the guidance of renal intervention and for the prediction of outcome.

Plasma creatinine (P-creatinine) is the most commonly used marker of glomerular filtration rate (GFR), which is normally used to assess the overall renal function [28, 29]. However, creatinine has a low sensitivity and is not a reliable marker for detecting early development of renal dysfunction [28]. Estimated creatinine clearance can be used to increase accuracy, but it is still based on plasma creatinine levels [28]. Cystatin C has been suggested as a more sensitive marker of GFR than creatinine. It detects even a small decline in renal function and has been well documented for several patient categories [30-34]. The use of cystatin C in cardiac surgery has been suggested in a few studies [35-40]. It has, however, not been validated as a marker of GFR and renal function post-operatively in cardiac surgery. Iohexol clearance is considered a reliable reference

method and gold standard for the determination of GFR and has previously been used to validate cystatin C [41, 42], but not in cardiac surgery.

The practice of retransfusing shed blood during cardiac surgery with cardiopulmonary bypass (CPB) using cardiotomy suction has been shown to be a major source of lipid microemboli (LME) [5, 6, 10, 43]. These lipid emboli form small occlusions in the vasculature, and histological examination of the brain after cardiac surgery has demonstrated lipid deposits in the capillaries in this organ [10]. These phenomenon found in the capillary bed is sometimes referred to as small capillary arteriolar dilatations (SCADS) [10]. Until now, attention has been focused on emboli in the brain and the effects they may have on cerebral function. The kinetics of LME in the circulation and the distribution of embolic load to different organs have so far not been fully investigated.

Cardiotomy suction

In the majority of cases, when open heart surgery is performed, CPB are used. The blood is usually led out of the body by a tube, connected to a cannula in the right atrium and the inferior caval vein, into a reservoir. From the reservoir the blood is pumped through an oxygenator, where the gas exchange is carried out. The oxygenated blood is then led back into the patient through a cannula in the aorta. With the patient fully anticoagulated with heparin, the surgeon has the option to use cardiotomy suction. It is a nozzle with tubing that is connected to the blood reservoir in the CPB machine. Through the nozzle, shed blood from the operation field can be sucked into the reservoir and then be retransfused back into the circulation of the patient.

Liquid scintillation process

Scintillation is used to detect radiation in a sample and convert the radiation into a photon, which can be counted. A tissue sample, in this case containing beta radiation, is dissolved with a solvent. The molecules originating from the solvent are by far more than the molecules originating from the tissue sample. This means that every molecule from the tissue sample is surrounded by molecules from the solvent. When the radioactive substance disintegrates, beta particles will be emitted. The particles collide with the solvent molecules and transmit energy to the solvent molecule, which will be excited and charged with energy. This energy will then be retransmitted to another nearby solvent molecule and the process will go on until a solvent molecule reaches a scintillation molecule. The scintillation molecule will be excited and charged with energy. The scintillator molecule will thereafter return to the ground state, resulting in the emission of a light photon. This light usually has an unfavourable wavelength for the detection

and transformation to an electrical signal. Therefore, a secondary scintillator is added. The light photon will be absorbed by a secondary scintillator molecule, which will be excited. It returns to the ground state, while emitting a new light photon. This new light photon will have a longer and more favourable wavelength than the one previously absorbed. The new light photon is then detected by a photo multiplication device. This device converts the light from every detected photon into an electrical signal, which is also amplified. The device counts the signals, which produces a reading of the radiation from the tissue sample [44].

Iohexol

Iohexol is a non-ionic X-ray contrast medium with a molecular mass of 821 Da. It is eliminated from plasma by glomerular filtration without metabolism by the kidneys [45, 46] and does not bind to plasma protein [47-49]. Iohexol is considered to be a ideal marker for determination of GFR [50].

Cystatin C

Cystatin C is a 13.3 kDa protein that is produced in all nucleus-bearing cells. Production is fairly constant, does not change with muscle mass and is only affected to a small degree by age and gender. The protein is filtrated in the renal glomeruli, absorbed into the cells of the tubules and fully metabolized there. Cystatin C has a higher sensitivity for GFR than creatinine does, and is therefore superior to creatinine for detecting early stages of renal dysfunction [28, 31, 32, 51, 52]. A few studies have also shown that cystatin C can predict early development of acute kidney injury [36, 37] and duration of stay in intensive care after cardiac surgery [36]. Cystatin C levels have been reported to be altered in patients with thyroid dysfunction or on glucocorticoid therapy [53, 54]. Cystatin C levels are higher in a hyperthyroid state and lower in a hypothyroid state, and the difference compared with the euthyroid state was reported to be in the magnitude of 30% [54]. In patients with impaired renal function receiving corticosteroids, especially methylprednisolone pulses, the increase of cystatin C is not proportional to GFR impairment [53]. Therefore, care should be taken when interpreting cystatin C in patients with known thyroid dysfunction and patients with newly transplanted organs. Cystatin C has repeatedly been documented as a reliable marker of GFR and renal function and is well suited for clinical use.

Creatinine

Creatinine, which is a metabolic end stage product of creatine, is the most commonly used marker of GFR and renal function. The major weakness with creatinine as a marker is the variation in its release into the blood. Creatinine production changes significantly depending on muscle mass, age and gender [28]. In addition, plasma creatinine levels are affected by protein intake, especially large quantities of boiled meat [28]. In parallel with cystatin C, creatinine is filtered by the glomeruli, but it is also secreted by the renal tubules. When GFR is normal, the secretion of creatinine by the renal tubules is rather insignificant. As GFR decreases, the tubular secretion of creatinine will increase, thereby increasing the influence of tubular secretion on creatinine clearance. In addition, some drugs can reduce tubular secretion and increase creatinine concentration without an actual reduction of GFR [28]. Thyroid dysfunction may also alter creatinine levels. They have been found to be increased in hypothyroidism and decreased in hyperthyroidism [54].

Aims of the Studies

Study I
To investigate the distribution of lipid microemboli to different organs after cardiac surgery in a novel porcine model.

Study II
To study the kinetics of lipid emboli in the systemic circulation and the renal circulation during cardiac surgery in a porcine model.

Study III
To evaluate plasma cystatin C with iohexol clearance in cardiac surgery. The study also compared the accuracy of estimated clearance based on cystatin C with estimated clearance based on creatinine in cardiac surgery.

Study IV
To evaluate whether routine outcome variables in cardiac surgery without cardiotomy suction differed from cardiac surgery with cardiotomy suction.

Material and Methods

Study protocol

Study I
After gaining approval from the Regional Animal Study Ethics Committee, 10 adult pigs were anaesthetized and underwent a sternotomy. The right atrium and the ascending aorta were cannulated after a full dose of heparin (LEO Pharma A/S, Copenhagen, Denmark) had been administered. Cardiopulmonary bypass was instituted. All animals underwent standardized perfusion for 40 minutes. A shed blood phantom was infused into the cardiotomy reservoir of the heart-lung machine after 20 minutes of bypass. In eight animals (called the radioactive group), the blood phantom contained beta radioactive tritium-labelled triolein. In the remaining two animals (called the nonradioactive group), the blood phantom did not contain any beta radioactive tritium-labelled triolein. Tissue samples were taken from the white and gray matter of the cerebrum, brainstem, hippocampus, cerebellum, heart, left lung, liver, the cortex of one kidney, spleen, small intestine and skeletal muscle. In all animals, four samples were taken from each organ. The tissue samples were dissolved, decolorized and the level of radioactivity was measured. All samples had their radioactivity measured twice by liquid scintillation.

Study II
After approval from the Regional Animal Study Ethics Committee, eleven adult pigs were anaesthetized and underwent a sternotomy. In seven animals (called the systemic group), the right atrium and the ascending aorta were cannulated after a full dose of heparin had been administered. Cardiopulmonary bypass was instituted, and the animals underwent standardized perfusion for 40 minutes. A shed blood phantom, containing beta radioactive tritium-labelled triolein, was infused into the cardiotomy reservoir of the heart-lung machine after 20 minutes of bypass, and blood was sampled from the femoral artery and the internal jugular vein. Urine samples were obtained from the urine bladder of each animal after sacrificing the animal.

The remaining four animals (called the renal group) underwent a laparotomy, but were not canullated or put on CPB. An identical blood phantom as above was infused in the ascending aorta, and blood was sampled from the renal artery and vein. The blood and urine samples were dissolved and decolorized and the level of radioactivity was measured. All samples had their radioactivity measured twice by liquid scintillation.

Study III

After approval by the local ethics committee, informed consent was obtained from 21 patients, who were prospectively enrolled in the study. All patients were scheduled for elective coronary artery bypass grafting (CABG). Exclusion criteria were known iohexol allergy, pre- or postoperative dialysis, a preoperative plasma creatinine above 150 mmol/l or a peroperative finding that required a change in surgical procedure. Iohexol clearance was determined preoperatively and on the second postoperative day (details in separate section about iohexol clearance protocol). Plasma creatinine, plasma cystatin C and plasma C- reactive protein (CRP) were determined preoperatively and on the first, second, third and fifth postoperative day. Fluid balance for the patients was estimated for the first 2 postoperative days including the operation itself. Estimated creatinine clearance was calculated according to the formula by Cockcroft–Gault (CG) [55] and according to the equation developed from the Modification of Diet in Renal Disease study data (MDRD) [56]. Estimated cystatin C clearance (eCyC) was calculated according to the formula by Grubb et al [57]. Patient characteristics, CPB time and aortic cross clamping time were extracted from patient journals and the in-house surgical database.

Study IV

After approval by the local ethics committee, informed consent was obtained from 150 patients, who where prospectively enrolled in the study. All patients were scheduled for elective CABG. The patients were divided in two equally large groups. Cardiotomy suction was used in the control group. In the intended study group, the use of cardiotomy suction was to be avoided. Exclusion criteria were pre- or postoperative dialysis or a peroperative finding that required a change in surgical procedure. Plasma creatinine (P-creatinine), plasma C-reactive protein (P-CRP), plasma cystatin C (P-cystatin C), blood hemoglobin (B-hemoglobin), blood leucocytes (B-leucocytes), blood platelets (B-platelets) and plasma urea (P-urea) were analyzed preoperatively, on the first, second, third and fifth postoperative day. Plasma aspartate-aminotranspherase (P-ASAT), plasma alanine-aminotranspherase (P-ALAT) and plasma creatine kinase subunit MB (P-CKMB) were analyzed preoperatively and on the first postoperative day. Peroperative fluid balance for the patients was estimated by recording all given fluids and all losses. Transfusion of erythrocytes, plasma and platelets were recorded, as was the use of norepinephrine. Patient characteristics,

postoperative ventilator time, CPB time, aortic cross clamping time, number of grafts, complications and postoperative bleeding were extracted from patient journals and the in-house surgical database.

Anaesthesia in laboratory study (study I, II)

Premedication was performed with an intramuscular injection of 15 mg/kg ketamine chloride (Ketalar; Pfizer, New York, NY) and 0.2 mg/kg xylasine (Rompun; Bayer, Gothenburg, Sweden). Anaesthesia was induced by an intravenous injection of sodium thiopental (Pentothal; Abbot, North Chicago, Illinois) 10 mg/kg and atropine (Atropin Merck NM; Merck NM, Stockholm, Sweden) 0.02 mg/kg. Surgical preparations were made for tracheotomy. After giving an intravenous injection of succinylcholine (Celocurin; Ipex, Solna, Sweden) 0.2 mg/kg to obtain muscle relaxation, the endotracheal tube was inserted. Anaesthesia was maintained by infusion of 0.15 mg $\cdot$ kg^{-1} $\cdot$ min^{-1} ketamine chloride and 0.01 mg $\cdot$ kg^{-1} $\cdot$ min^{-1} pancuronom bromide (Pavulon; NV Organon, Oss, Netherlands), or by infusion of 0.1 to 0.2 mg $\cdot$ kg^{-1} $\cdot$ min^{-1} propofol (Diprivan; Astra-Zeneca, Luton, United Kingdom) together with intermittent injections of fentanyl (Leptanal; Lilly, France) and atracuriumbesylate (Janssen-Cilag AB, Sollentuna, Sweden). The animals were connected to a ventilator and the pCO_2 was maintained at between 4.5 and 6.0 kPa.

Anaesthesia in clinical study (study III, IV)

Anaesthesia was induced by fentanyl (Fentanyl; B. Braun Melzungen AG, Melzungen, Germany), midazolam (Dormicum; F. Hoffmann-La Roche Ltd., Basel, Switzerland), propofol (Propofol-Lipuro; B. Braun Melzungen AG) and suxametonium (Celocurin; Ipex Medical AB, Solna, Sweden). Anaesthesia was maintained by propofol, fentanyl and vercuronium bromid (Norcuron; Schering-Plough AB, Stockholm, Sweden). In a few cases, isoflurane (Foren; Abbott Laboratories, Chicago, IL) was added. Dobutamine (Dobutamine; Hospira Inc., Lake Forest, IL) was used as the primary inotropic drug, and norepinephrine (Noradrenalin APL; Apoteket AB, Stockholm, Sweden) was used for vasoconstriction.

Administration of radioactive shed blood (study I, II)

To mimic mediastinal shed blood, in the aspect of lipids, triolein was used. Triolein has been used in several studies before with aim to study lipid embolization to lung and brain [58-61]. In addition, triolein consists of three chains of oleic acid, which is the most common fatty acid in human adipose tissue [62] (Table 1). A solution of radioactive triolein was prepared by mixing a 65% nonradioactive triolein solution (Carl Roth GmbH, Karlsruhe, Germany) with radioactive tritium-labeled triolein (Amersham BioSciences, Little Chalfont, United Kingdom). The proportions used were such that 5 mL of the final solution should contain 1 mCi of radioactivity. A shed blood phantom was made by mixing 200 mL blood from the cardiotomy reservoir with 200 mL saline and 5 mL radioactive triolein solution.

Table 1. The most common free fatty acids in human adipocytes.

Free fatty acid	C:Double Bonds	Proportion
Oleic	18:1	49.9%
Palmatic	16:0	23.0%
Linolic	18:2	9.5%
Palmitoleic	16:1	5.8%
Myristic	14:0	2.9%

The middle column gives the composition in terms of the number of carbon atoms per molecule and double bond content (after the colon).

Processing of samples in laboratory study (study I, II)

The tissue samples in study I and the blood/urine samples in study II were dissolved and decolorized with Soluene 350 (Packard Bioscience, Groningen, Netherlands) and hydrogen peroxide respectively. Scintillation fluid (Hionic Fluor; Packard Bioscience) was added and in study II also 95% ethanol. The level of radioactivity (beta radiation) was measured by scintillation counting, using a liquid scintillation counter (14814 Win Spectral Guardian; Wallac Oy, Turku, Finland) together with the software supplied by the manufacturer (study I, Easy Count; study II, Tritium Count). The beta radiation from the tritium-

labelled triolein was used as a marker for triolein content. Radioactivity was reported as the number of disintegrations per minute per gram sample of tissue (DPM/g) in study I and as the number of disintegrations per minute per ml sample of blood/urine (DPM/ml) in study II. The rationale for not using the same software in study I and II was that Easy Count measures tissue samples better, whereas Tritium Count is better in measuring blood samples.

Iohexol clearance protocol (study III)

A preoperative blood sample (baseline test) was drawn from a peripheral vein to determine whether any residual iohexol was present in the blood. The patient then received an intravenous injection of 5 ml iohexol (Omnipaque 300mgI/mL; GE Healthcare, Oslo, Norway) in the same venous line. Blood samples were taken from a contra lateral peripheral vein line 180 and 240 min after the injection. The second postoperative day, baseline testing, iohexol injection and blood sampling were performed in a manner identical to the preoperative sampling, with the exception of vein access, which was obtained through a double-lumen central venous catheter. Serum iohexol (S-iohexol) concentration was determined using a high-pressure liquid chromatography method [48]. A two-point plasma clearance of iohexol was calculated assuming a one-compartment model for the distribution of iohexol. Slope clearance was calculated according to the formula by Bröchner-Mortensen [63].

Calculations

Iohexol clearance formula: [63, 88]

Clearance = CIBMkorr x (1.73/BSA)

$CIBMKorr = 0.99078 \times Cl1 - 0.001218 \times Cl1^2$

$Cl1 = Dose \times 1000 \times Slope / e^{intercept}$

$Intercept = Iohexol_{T1} + Slope \times T1$

$Slope = [Ln(Iohexol_{T1}) - Ln(Iohexol_{T2})] / (T2-T1)$

$BSA = 0.007184 \times weight\ (kg)^{0.425} \times height\ (cm)^{0.725}$

T1 = time for first iohexol sample (minutes)
T2 = time for second iohexol sample (minutes)
$Iohexol_{T1}$ = Iohexol level at T1 (mg/L)
$Iohexol_{T2}$ = Iohexol level at T2 (mg/L)
Dose = Amount iohexol (milligrams)
BSA= Body Surface Area (m^2) [89]

Estimation of glomerular filtration rate (GFR) based on plasma cystatin C:
Equation by Grubb [57]:
GFR (mL/min/1.73m^2) = 84.69 x cystatin C (mg/L)$^{-1.680}$ x (1.384 if a child is < 14 years old) x (0,948 if females)

Estimation of glomerular filtration rate (GFR) based on creatinine:
Equation by Cockcroft-Gault [55]:
GFR mL/min = (140-age) x weight kg / plasma creatinine µmol/L x Constant
Constant is 1.23 for men and 1.04 for women

MDRD study equation [56]:
GFR mL/min/1.73 m^2 = 186 x (plasma creatinine µmol/L/88.4)$^{-1.154}$ x (age)$^{-0.203}$ x (0.742 if female) x (1.210 if Afro-American)

Statistical analysis

The Statistica software package versions 6-8 (Statsoft, Tulsa, Ok, USA) was used for all statistics in study I - IV.

Study I
For each organ, a mean value was determined from the four samples and the two radioactivity measurements. Values for the control animals and the case group were expressed as mean ± 1 SD, unless otherwise stated. In addition, the radioactivity in each animal was standardized by presenting the amount of radioactivity found in each organ or tissue as a percentage of the total measured radioactivity in the animal (sum of all examined organs or tissues). To compare radioactivity between different tissues, a Student´s t-test was performed. To compare differences in radioactivity within different regions of the brain, a repeated measurement analysis of variance analysis (ANOVA) was performed. A p-value < 0.05 was considered significant.

Study II
For each blood sample, a mean value was determined from the two radioactivity measurements. In both the systemic and renal group, the area under curve (AUC) was calculated from the start of infusion to 20 and 15 minutes respectively after the start of infusion. Background radiation (determined as the mean of the preinfusion sample and the sample taken at the start of the infusion) was subtracted from the levels used for the calculation of the AUC. For comparison of radioactivity between sampling intervals a Wilcoxon-test was performed. Unless otherwise stated, the distribution was expressed as the median and the inter quartile range between first and third quartile. A p-value < 0.05 was considered significant.

Study III

Univariate and multivariate regression analysis was performed and correlation coefficients were determined. For repeated measures, an ANOVA analysis was performed first, and if significant, a Student's t-test for dependent samples was used to compare preoperative and postoperative data to determine which variable differed from preoperative measurements. If not otherwise stated, all measurements were presented as mean ± 1 SD. A p-value < 0.05 was considered significant.

Study IV

A Student´s t-test for independent samples was used to compare data from the different groups. When data were skewed distributed, a Mann-Whitney U-test was used. If not otherwise stated, all measurements were presented as mean ± 1 SD. A p-value < 0.05 was considered significant.

Results

Study I

No beta radiation was detected in the tissues of the nonradioactive animals (Table 2). In the radioactive group, beta radiation was detected in all tissues examined indicating the presence of LME. The highest levels of radioactivity were found in samples taken from the kidney and spleen. The radioactivity in these organs was approximately 5 to 10 times higher than in the other organs examined (Table 2). Liver and the gray matter of the cerebrum showed the third and fourth highest levels of beta radiation, respectively. The lowest levels were found in skeletal muscle. No radiation was detected in venous blood before the shed the blood phantom was added to the circulation. High levels of radioactivity were found in the shed blood phantom (Table 2). A high variation in radioactivity from animal to animal was found. However, the relative variation, between the organs in the animals was low (Figure 1). Tissue levels were compared with an aggregated mean value from all brain regions. This comparison revealed that the kidney and spleen had higher levels of beta radiation than the brain, and that muscle had lower levels of beta radiation than the brain (Table 2). The distribution of beta radiation in the regions of the brain examined showed differences that were statistically significant when tested with ANOVA analysis (Table 2). The level of radioactivity found in gray matter of cerebrum was significantly higher than in the other regions of the brain examined (Student`s t-test, $p < 0.05$ in all tests). The lowest levels of radiation within the brain were found in the brainstem. No radiation was detected in venous blood before the animals were sacrificed.

Table 2. Beta radiation in different organs.

Tissue	Nonradioactive Animal	Radioactive Animals	
Brain [a]		11 797 ± 10 311	
White matter cerebrum	0 ± 0	10 188 ± 9 553	p<0.05 [b]
Gray matter cerebrum	0 ± 0	18 356 ± 14 550	
Brainstem	0 ± 0	7 054 ± 6 142	
Hippocampus	0 ± 0	11 412 ± 13 034	
Cerebellum	0 ± 0	12 777 ± 10 343	
Tissue			
Heart	0 ± 0	11 107 ± 11 460	n.s [c]
Lung	0 ± 0	17 117 ± 24 817	n.s [c]
Liver	0 ± 0	25 614 ± 1 8342	n.s [c]
Kidney	0 ± 0	176 385 ± 118 563	p<0.005 [c]
Spleen	0 ± 0	139 565 ± 138 274	p<0.05 [c]
Small Intestine	0 ± 0	17 332 ± 12 910	n.s.[c]
Muscle	0 ± 0	1 980 ± 1 879	p<0.05 [c]
Venous blood			
Before infusion		0 ± 0	
At euthanasia		0 ± 0	
Shed blood phantom		1 517 416 ± 1 299 906	

Beta radiation expressed as disintegrations per minute per gram of tissue (DPM/g) or milliliters of blood. [a] - Mean brain was calculated as a mean of the different brain areas sample. [b] - Repeated measurement ANOVA showed a significant difference within the different regions of the brain. [c] - Tissue levels were compared with mean brain levels with a Student´s t-test.

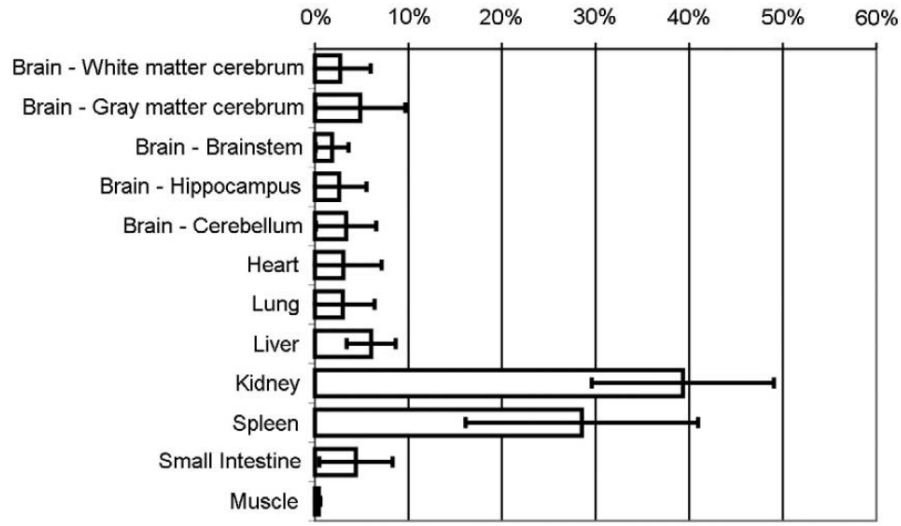

Figure 1.

Relative distribution of radioactivity, shown as the standardized concentration of lipid microemboli in different tissues (levels are expressed as a percent of total radioactivity from all tissue examined for each animal).

Study II

After infusion of the radioactive shed blood phantom in the systemic group, a median of 130 (32-212)-fold increase in radioactivity was observed in arterial samples between pre-infusion levels and peak level (Figure 2). The median time from starting infusion to observation of peak levels in arterial samples was 40 (40-60) seconds. In six of seven animals a second peak of radioactivity was observed in arterial blood and in three animals also a third peak. In the venous samples, an 18 (5-26)-fold increase in radioactivity was observed for the first peak, which was observed at a median time of 60 (60-80) seconds after the start of the infusion. At the end of the experiment, the radioactivity was slightly increased (Figure 2) in arterial blood as well as in the venous blood compared to the levels before infusion of shed blood. In the systemic group, the ratio of the AUC in arterial blood and in venous blood was 0.381 (0.173-0.437), ($p < 0.02$), i.e. 38% of the radioactivity found in arterial blood was also found on the venous side. When AUC was calculated for the first three minutes the ratio between arterial and venous blood was 0.179 (0.086-0.315). After infusion of shed blood, the blood pressure decreased in all animals (Figure 3) The blood pressure was thereafter spontaneously restored. The radioactivity in urine were overall low at the end of the experiment, and were about the same level as the radioactivity found in blood before the start of infusion of radioactive shed

blood (Table 3). After infusion of the radioactive shed blood phantom in the renal group, a median 27-fold increase in radioactivity, compared to background radiation, was observed in samples taken from the renal artery. The peak levels were observed at a median time of 3 (2-4) minutes after the start of the infusion and then the radioactivity dropped fast and was levelled out in all animals at the latest 6 minutes after the start of the infusion. No second peak of radioactivity was observed in any animal. In the samples taken from the renal vein, a median 4.8-fold increase in radioactivity was observed with a median peak 4 (2.75-5) minutes after the start of the infusion. At the end of the experiment the median radioactivity in the renal artery and vein were slightly increased compared to the levels found before the start of the infusion (Figure 4). In the renal group, the ratio of the 15 minute AUC in the renal artery and the renal vein was 0.229 (0.081-0.350), i.e. 23% of the radioactivity found on the renal artery was also found in the renal vein.

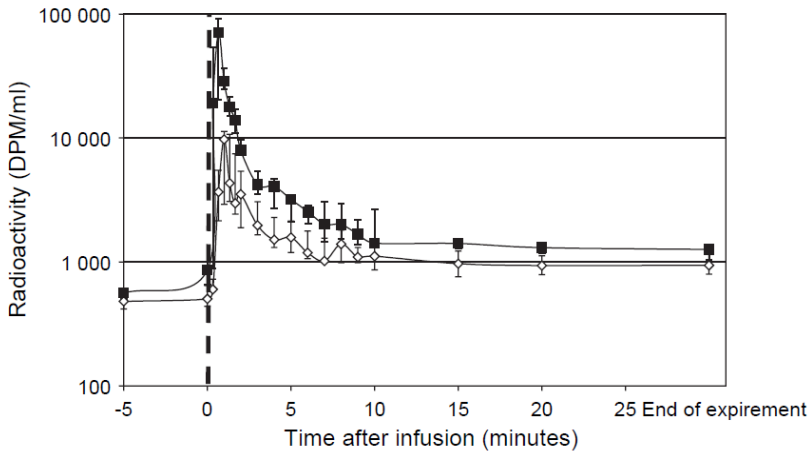

Figure 2.

Median radioactivity in arterial (black squares) and venous (white diamonds) blood during the experiments, expressed as median (IQR) for animals in the systemic group. The dotted line indicates the time at which the shed blood was infused. The last observation represents the end of the experiment when the animals were sacrificed. Abbreviations: DPM/ml, disintegrations per minute per milliliter; IQR, inter quartile range.

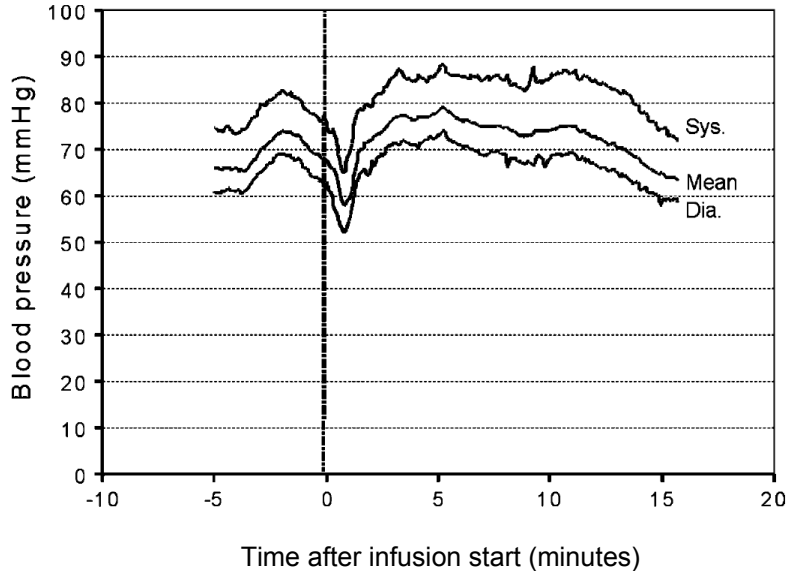

Figure 3.

Mean arterial pressure (systolic, mean and diastolic) for animals in the systemic group during a period 5 minutes prior to infusion and 20 minutes thereafter. The dotted line indicates the time at which the shed blood was infused.

Table 3.

Beta radiation in blood and urine.

Animal no.	AUC Art (DPM/ml x min)	AUC Ven (DPM/ml x min)	First passage	Urine (DPM/ml)
1	75 767	30 798	59.4%	78
2	105 900	19 918	81.2%	278
3	109 520	14 207	87.0%	657
4	235 905	89 831	61.9%	161
5	50 313	23 539	53.2%	118
6	51 088	35 741	30.0%	153
7	75 823	12 001	84.2%	82
Median	75 823	23 539	61.9%	153
(IQR)	(63 449 – 107 724)	(17 094 – 33 270)		(100 – 220)

Abbreviations: AUC, area under curve; DPM/ml, disintegrations per minute per milliliter; IQR, inter quartile range; min, minute.

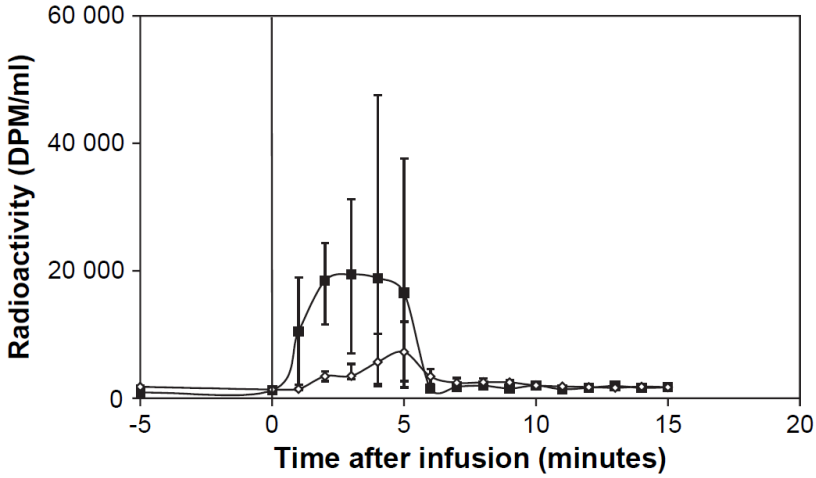

Figure 4.

Median radioactivity in the renal artery (black squares) and the renal vein (white diamonds) from animals in the renal group. The vertical line indicates the start of the infusion of shed blood. Abbreviations: DPM/ml, disintegrations per minute per milliliter.

Study III

One patient was excluded because of postoperative dialysis. The baseline characteristics for the study patients are summarized in Table 4. The concentration of iohexol before injection was zero in all patients before and after surgery. The concentrations of S-iohexol, P-cystatin C, P-creatinine, P-CRP, iohexol clearance and creatinine clearance (CG and MDRD) preoperatively and on the second postoperative day are displayed in Table 5. Preoperative iohexol clearance correlated to preoperative P-cystatin C ($r = -0.80$, $p < 0.0001$), P-creatinine ($r = -0.74$, $p < 0.005$) and creatinine clearance [$r = 0.63$ (CG) and $r = 0.70$ (MDRD), both $p < 0.005$].

Postoperative iohexol clearance correlated to postoperative P-cystatin C, P-creatinine and creatinine clearance. The strongest correlation to iohexol clearance was found with P-cystatin C (Table 6). Eight patients had a decreased postoperative renal function (postoperative iohexol clearance < 90 ml/min/1.73m^2) and a subgroup analysis was performed in this group. The strongest correlation to iohexol clearance was found with P-cystatin C (Table 6). Cystatin C clearance (eCyC) and creatinine clearance (CG and MDRD) were compared with iohexol clearance. A significant difference was found between iohexol clearance and both clearances based on creatinine (CG and MDRD). Cystatin C clearance (eCyC) did not differ from iohexol clearance

(Table 7). No correlation was found between P-cystatin C and P-CRP. On the first postoperative day, both P-cystatin C and P-creatinine declined significantly compared with their preoperative baseline level. On the second postoperative day, P-creatinine concentration returned to the preoperative baseline level and remained constant through out the study time. P-cystatin C was significantly increased on the second postoperative day compared with the preoperative baseline level. It remained at a significantly higher level compared with baseline level on the third postoperative day and thereafter it returned to the baseline level on the fifth postoperative day. The total fluid balance was +4348 ± 1387 ml on the day of surgery, and +3646 ± 1821 ml and +3224 ± 1874 ml on the first and second postoperative day, respectively.

Table 4.
Baseline characteristics of the patients.

Variable (n = 20)	**Mean ± SD or n(%)**
Age (years)	69.0 ± 9.8
Male, number (%)	17 (85%)
LVEF > 50%	13 (65%)
LVEF 50-30%	5 (25%)
LVEF < 30%	2 (10%)
Preoperative Euroscore	4.4 ± 2.4
Diabetes, number (%)	5 (25%)
CCS angina grade	2.4 ± 0.6
NYHA heart failure grade	1.7 ± 0.7
CPB time (min)	72.4 ± 21.2
Aortic cross-clamp time (min)	45.0 ± 16.1

Abbreviations: SD, standard deviation; LVEF, left ventricular ejection fraction; CCS, Canadian Cardiovascular Society Class; NYHA, New York Heart Association Class; CPB, cardiopulmonary bypass.

Table 5.
Concentrations and clearance estimations for the entire study group.

	Preoperative	Postoperative day two	p
n=20			
P-cystatin C (mg/L)	1.02 ± 0.33	1.21 ± 0.44	< 0.005
P-creatinine (µmol/L)	78.3 ± 15.8	80.1 ± 24.9	n.s
P-CRP (mg/L)	12.5 ± 16.5	184.1 ± 50.4	< 0.0001
S-iohexol 180 min after injection (mg/L)	62.0 ± 21.8	57.8 ± 23.8	n.s
S-iohexol 240 min after injection (mg/L)	47.2 ± 22.9	46.3 ± 24.5	n.s
iohexol clearance (mL/min/1.73 m^2)	81.4 ± 22.0	83.4 ± 29.9	n.s
creatinine clearance (CG) (mL/min)	91.2 ± 23.9	98.1 ± 32.4	n.s
creatinine clearance (MDRD) (mL/min/1.73 m^2)	85.3 ± 17.0	87.6 ± 25.7	n.s

Abbreviations: CRP, C-reactive protein; CG, Cockcroft-Gault formula; MDRD, the modification of diet in renal disease study formula.

Table 6.
Correlation between postoperative iohexol clearance and postoperative measures of renal function.

A n = 20	**r**	**r^2**	**p**
cystatin C	-0.9	0.81	<0.0001
1/ cystatin C	0.86	0.74	<0.0001
creatinine	-0.83	0.7	<0.0001
1/creatinine	0.78	0.61	<0.0001
creatinine clearance (CG)	0.82	0.67	<0.0001
creatinine clearance (MDRD)	0.85	0.74	<0.0001
B n = 8			
cystatin C	-0.99	0.98	<0.0001
1/ cystatin C	0.97	0.94	<0.0001
creatinine	-0.88	0.78	<0.005
1/creatinine	0.95	0.91	<0.0005
creatinine clearance (CG)	0.88	0.78	<0.005
creatinine clearance (MDRD)	0.95	0.9	<0.0005
cystatin C clearance (eCyC)	0.95	0.90	<0.0005

A = entire group. B = a subgroup with postoperative iohexol clearance < 90 ml/min/1.73 m^2. Postoperative measures refer to the second postoperative day. Abbreviations: r, correlation coefficient; CG, Cockcroft-Gault formula; MDRD, the modification of diet in renal disease study formula; eCyC, estimated cystatin C clearance.

Table 7.
Postoperative clearance calculations.

Clearance n=8	Mean	± SD	p
iohexol clearance	53.5	23.4	
creatinine clearance (CG)	75.8	32.3	0.006
creatinine clearance (MDRD)	70.0	22.9	0.0005
cystatin C clearance (eCyC)	54.5	30.4	0.81

Postoperative clearance calculated by iohexol injection compared with three different methods for estimating renal clearance (GFR) in patients with a postoperative iohexol clearance < 90 ml/min/1.73 m^2. Measures are made on the second postoperative day. Creatinine clearance (CG) in ml/min. All other clearances in ml/min/1,73 m^2. Abbreviations: CG, Cockcroft-Gault formula; MDRD, the modification of diet in renal disease study formula; eCyC, estimated cystatin C clearance.

Study IV

For the analysis, the study cohort was divided in four different groups. First, the control group (n=71) in which cardiotomy suction was used. Secondly, the intended study group (n=75) in which the intention from start was to not use cardiotomy suction. In addition, a cross-over group (X-over group) was formed (n=23), in which, against the former intention, the use of cardiotomy suction was inevitable because of peroperative bleeding. Finally, the study group (n=52), which included the remaining patients, where cardiotomy suction was not used (figure 5). One patient was excluded because of postoperative dialysis and one patient was excluded because of change of surgical procedure. Two patients were excluded because of incomplete data. All excluded patients belonged to the control group.

The intended study group revealed a higher degree of effort angina according to the Canadian Cardiovascular Society grading scale (CCS) compared with the control group preoperatively (table 8). Otherwise the two groups were comparable without further significant differences. The control group and the study group differed in two preoperative variables. The age and the CCS score were significantly higher in the study group compared with the control group (table 8).

In outcome variables, the amount of transfused packed red blood cell units during surgery, and also throughout the whole study period, was significantly larger in the control group compared with the study group (table 9). There was a tendency for a larger amount of transfused plasma units in the control group

throughout the study period compared with the study group. If the 23 patients in the X-over group (in which cardiotomy suction also was used during surgery) were added to the control group, the difference in transfused units of plasma became significant (table 9). The number of patients, who received transfusion of plasma throughout the study period, was significantly higher in the control group compared with the study group (table 9). Postoperative P-ASAT and P-ALAT were significantly higher in the control group compared with the study group (table 10). The control group had a significantly longer CPB time and aortic clamping time compared with the study group (table 11). Diuresis and the use of norepinephrin during surgery were significantly higher in the control group compared with the study group. The same was true for peroperative fluid balance if the X-over group was added to the control group (table 11).

In an intentions-to-treat analysis, outcome variables were compared between the control group and the intended study group (table 12-14) and between the study group and the X-over group (table 15-18). To summarize the significant findings of this analysis, postoperative ASAT and ALAT were significantly higher in the control group compared with the intended study group, and so was the aortic clamping time. Transfusion of red blood cell units, plasma units and platelet units extended over the whole study period were significantly higher in the X-over group compared with the study group as well as the percentage of patients that received plasma and platelet transfusion. Peroperative fluid balance, peroperative diuresis, preoperative norepinephrine consumption and CPB time were significantly higher in the X-over group compared with the study group, and so was also preoperative CCS score.

There were no significant differences between the control group and the study group in the following variables: B-hemoglobin, B-platelets, B-leucocytes, P-CRP, P-cystatin C, P-creatinine, P-urea, P-CKMB, number of grafts, postoperative bleeding, reoperation due to bleeding, postoperative ventilator time, postoperative ICU stay, postoperative use of norepinephrine or inotropics, postoperative infection, transfusion of platelet units or percent of platelet recipients, and finally, percent of red blood cell transfusion recipients.

There was no thirty-day mortality in this study. The one-year mortality amounted to one patient in the control group and one patient in the study group.

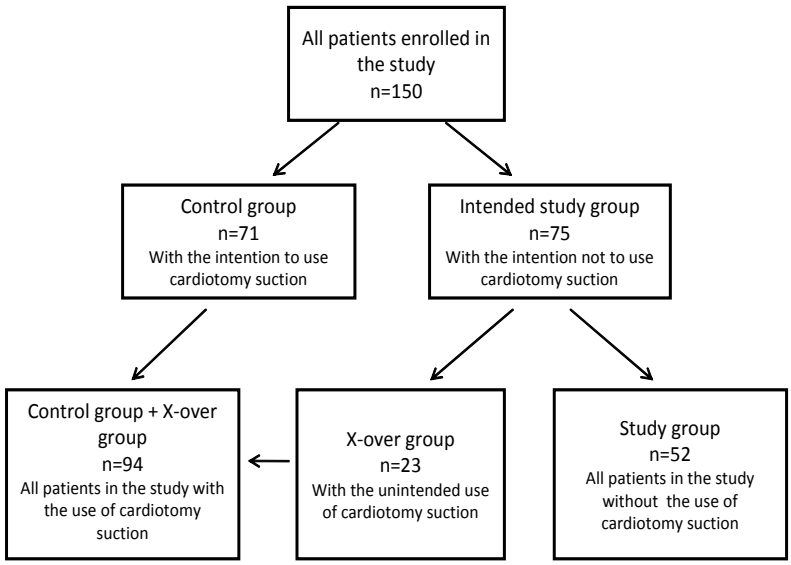

Figure 5.
Groups in the study.

Table 8.
Baseline characteristics.

	Control group	Intended study group	p	Study group	p
n	71	75		52	
Age	66.8 ± 8.9	69.8 ± 9.9	n.s	70.3 ± 9.7	< 0.05
Female gender n (%)	16 (22.5)	14 (18.7)	n.s	7 (13.5)	n.s
Preoperative Euroscore	4.4 ± 3.1	4.6 ± 2.5	n.s	4.2 ± 2.4	n.s
Diabetes n (%)	15(21.1)	18 (24.0)	n.s	12 (23.1)	n.s
COPD n (%)	7(9.8)	5 (6.7)	n.s	2 (3.8)	n.s
LVEF > 50% n (%)	47 (66.2)	50 (66.7)	n.s	38 (73.1)	n.s
LVEF 30-50% n (%)	18 (25.3)	21(28.0)	n.s	12 (23.1)	n.s
LVEF < 30% n (%)	6 (8.4)	4 (5.3)	n.s	2 (3.8)	n.s
NYHA class					
I-II n (%)	52 (73.2)	49 (65.3)	n.s	36 (69.2)	n.s
III-IV n (%)	19 (26.8)	26 (34.7)	n.s	16 (30.8)	n.s
CCS class					
I-II n (%)	45 (63.4)	19 (25.3)	< 0.0001	17 (32.7)	< 0.005
III-IV n (%)	26 (36.6)	56 (74.7)	< 0.0001	35 (67.3)	< 0.005

Control group – with the intention to use cardiotomy suction. Intended study group – group with the intention not to use cardiotomy suction. Study group – group without the use of cardiotomy suction. Values are expressed as mean ± 1 standard deviation or number (percent). All comparisons are made against control group. Abbreviations: COPD, chronic obstructive pulmonary disease; LVEF, left ventricular ejection fraction; NYHA, New York Heart Association class; CCS, Canadian Cardiovascular Society class; n.s, no significant difference.

Table 9.
Transfusion of blood products and B-hemoglobin analysis.

	Study group	Control group	p	Control group with X-over group	p
n	52	71		94	
Hemoglobin preop (g/L)	139.0 ± 13.3	135.8 ± 12.8	n.s	136.0 ± 12.9	n.s
Hemoglobin postop max (g/L)	106.8 ± 10.9	109.0 ± 9.1	n.s	107.8 ± 9.0	n.s
Hemoglobin postop min (g/L)	91.9 ± 10.2	90.0 ± 13.9	n.s	89.4 ± 13.0	n.s
Peroperative					
Received red blood cell transfusion n (%)	7 (13.5)	18 (25.4)	n.s	24 (25.5)	n.s
Transfused red blood cells units	0.19 ± 0.53	0.66 ± 1.64	< 0.05	0.64 ± 1.56	< 0.05
Received plasma transfusion n (%)	1 (1.9)	5 (7.0)	n.s	7 (7.4)	n.s
Transfused plasma units	0.04 ± 0.28	0.17 ± 0.80	n.s	0.2 ± 0.83	n.s
Received platelet transfusion n (%)	3 (5.8)	2 (2.8)	n.s	3 (3.2)	n.s
Transfused platelet units	0.10 ± 0.41	0.06 ± 0.34	n.s	0.06 ± 0.36	n.s
Extended over the whole study period					
Received red blood cell transfusion n (%)	23 (44.2)	42 (59.2)	n.s	57 (60.6)	n.s
Transfused red blood cells units	1.12 ± 1.50	2.14 ± 2.88	< 0.05	2.18 ± 2.75	< 0.02
Received plasma transfusion n (%)	6 (11.5)	20 (28.2)	< 0.05	29 (30.8)	< 0.02
Transfused plasma units	0.44 ± 1.54	1.38 ± 3.36	n.s	1.54 ± 3.52	< 0.05
Received platelet transfusion n (%)	3 (5.8)	4 (5.6)	n.s	10 (10.6)	n.s
Transfused platelet units	0.10 ± 0.41	0.16 ± 0.71	n.s	0.27 ± 0.88	n.s

Study group – group without the use of cardiotomy suction. Control group – group with the use of cardiotomy suction. X-over group – group with the intention not to use cardiotomy suction, but where it had to be used because of blood loss. Values are expressed as mean ± 1 standard deviation or number (percent). All comparisons are made against study group. Abbrevaitions: preop, preoperative; postop max, maximum value recorded postoperatively; postop min, minimum value recorded postoperatively; n.s, no significant difference.

Table 10.
Laboratory analysis.

	Study group	Control group	p	Control group with X-over group	p
n	52	71		94	
ASAT preop (μkat/L)	0.50 ± 0.19	0.54 ± 0.27	n.s	0.53 ± 0.26	n.s
ASAT postop max (μkat/L)	0.63 ± 0.24	1.00 ± 0.93	< 0.01	0.95 ± 0.84	< 0.01
ALAT preop (μkat/L)	0.57 ± 0.33	0.70 ± 0.57	n.s	0.67 ± 0.54	n.s
ALAT postop max (μkat/L)	0.44 ± 0.20	0.57 ± 0.32	< 0.01	0.55 ± 0.34	< 0.05
CKMB preop (μg/L)	4.10 ± 1.85	4.01 ± 1.61	n.s	4.11 ± 2.09	n.s
CKMB postop max (μg/L)	15.82 ± 18.85	27.69 ± 45.86	n.s	25.70 ± 41.52	n.s
CRP preop (mg/L)	8.9 ± 13.8	8.08 ± 12.5	n.s	8.0 ± 11.6	n.s
CRP postop max (mg/L)	213.7 ± 73.5	214.6 ± 65.6	n.s	216.0 ± 69.4	n.s
Leucocytes preop (10^9/L)	7.47 ± 1.71	7.73 ± 1.88	n.s	7.64 ± 1.8	n.s
Leucocytes postop max (10^9/L)	13.0 ± 3.3	12.6 ± 3.22	n.s	12.3 ± 3.2	n.s
Platelets preop (10^9/L)	235.4 ± 69.3	243.1 ± 78.1	n.s	244.8 ± 78.5	n.s
Platelets postop max (10^9/L)	275.3 ± 151.3	254.0 ± 74.9	n.s	256.7 ± 82.1	n.s
Cystatin C preop (mg/L)	1.18 ± 0.42	1.14 ± 0.42	n.s	1.13 ± 0.38	n.s
Cystatin C postop max (mg/L)	1.46 ± 0.69	1.42 ± 0.64	n.s	1.40 ± 0.58	n.s
Creatinine preop (μmol/L)	89.4 ± 29.0	85.9 ± 27.8	n.s	84.6 ± 25.7	n.s
Creatinine postop max (μmol/L)	109.4 ± 53.8	100.4 ± 48.3	n.s	97.3 ± 43.7	n.s
Urea preop (mmol/L)	7.30 ± 2.73	7.39± 3.44	n.s	7.17 ± 3.15	n.s
Urea postop max (mmol/L)	8.64 ± 4.76	9.07 ± 8.74	n.s	8.58 ± 7.74	n.s

Study group – group without the use of cardiotomy suction. Control group – group with the use of cardiotomy suction. X-over group – group with the intention not to use cardiotomy suction, but where it had to be used because of blood loss. Values are expressed as mean ± 1 standard deviation or number (percent). All comparisons are made against study group. Abbreviations: preop, preoperative; postop max, maximum value recorded postoperatively; n.s, no significant difference.

Table 11.
Peroperative and postoperative variables.

	Study group	Control group	p	Control group with X-over group	p
n	52	71		94	
CPB time, minutes	69.5 ± 19.5	81.8 ± 25.1	< 0.005	82.4 ± 26.4	< 0.005
Aortic cross-clamping time minutes	42.8 ± 13.2	49.5 ± 15.4	< 0.02	48.2 ± 16.6	< 0.05
Number of grafts	2.73 ± 0.564	2.61 ± 0.644	n.s	2.66 ± 0.651	n.s
Peroperative fluid balance ml	2360.6 ± 879.0	3582.9 ± 1186.8	n.s	3683.0 ± 1307.7	< 0.05
Peroperative diuresis ml	534.6 ± 225.3	689.9 ± 399.2	< 0.02	711.6 ± 393.2	< 0.005
Peroperative norepinephrine µg	395 ± 344	650 ± 723	< 0.05	679 ± 873	< 0.05
Postoperative norepinephrine µg	1648 ± 1678	2100 ± 2890	n.s [a]	2041 ± 2720	n.s [a]
Postoperative ventilator time hours	6.88 ± 3.08	8.40 ± 9.60	n.s [a]	10.08 ± 15.71	n.s [a]
Postoperative bleeding ml	701.9 ± 467.0	850.4 ± 697.3	n.s	864.7 ± 687.0	n.s
Postoperative infektion no (%)	2 (3.8)	2 (2.8)	n.s	3 (3.2)	n.s
Postoperative inotropics >24 h no (%)	2 (3.8)	5 (7.0)	n.s	8 (8.5)	n.s
Postoperative ICU time hours	23.30 ± 8.36	29.51 ± 22.66	n.s [a]	30.23 ± 24.72	n.s [a]
Reoperation due to bleeding no (%)	2 (3.8)	1 (1.4)	n.s	1 (1.1)	n.s

Study group – group without the use of cardiotomy suction. Control group – group with the use of cardiotomy suction. X-over group – group with the intention not to use cardiotomy suction, but where it had to be used because of blood loss. Values are expressed as mean ± 1 standard deviation or number (percent). [a] - comparison is made with Mann-Whitney U-test. All comparisons are made against study group. Abbreviations: CPB, cardiopulmonary bypass; ICU, intensive care unit; n.s, no significant difference.

Table 12.
Transfusion of blood products and B-hemoglobin analysis.

	Control group	Intended study group	p
n	71	75	
Hemoglobin preop (g/L)	135.8 ± 12.8	138.3 ± 13.3	n.s
Hemoglobin postop max (g/L)	109.0 ± 9.1	106.0 ± 10.1	n.s
Hemoglobin postop min (g/L)	90.0 ± 13.9	90.5 ± 10.15	n.s
Peroperative			
Received red blood cell transfusion n (%)	18 (25.4)	13 (17.3)	n.s
Transfused red blood cells units	0.66 ± 1.64	0.31 ± 0.87	n.s
Received plasma transfusion n (%)	5 (7.0)	3 (4.0)	n.s
Transfused plasma units	0.17 ± 0.80	0.11 ± 0.56	n.s
Received platelet transfusion n (%)	2 (2.8)	4 (5.3)	n.s
Transfused platelet units	0.06 ± 0.34	0.09 ± 0.41	n.s
Extended over the whole study period			
Received red blood cell transfusion n (%)	42 (59.2)	38 (50.7)	n.s
Transfused red blood cells units	2.14 ± 2.88	1.48 ± 1.88	n.s
Received plasma transfusion n (%)	20 (28.2)	15 (20.0)	n.s
Transfused plasma units	1.38 ± 3.36	0.93 ± 2.64	n.s
Received platelet transfusion n (%)	4 (5.6)	9 (12.0)	n.s
Transfused platelet units	0.15 ± 0.71	0.25 ± 0.79	n.s

Control group – group with the use of cardiotomy suction. Intended study group – group with the intention not to use cardiotomy suction. Values are expressed as mean ± 1 standard deviation or number (percent). Abbrevaitions: preop, preoperative; postop max, maximum value recorded postoperatively; postop min, minimum value recorded postoperatively; n.s, no significant difference.

Table 13.
Laboratory analysis.

	Control group	Intended study group	p
n	71	75	
ASAT preop (µkat/L)	0.54 ± 0.27	0.50 ± 0.20	n.s
ASAT postop max (µkat/L)	1.00 ± 0.93	0.67 ± 0.33	p < 0.005
ALAT preop (µkat/L)	0.70 ± 0.57	0.57 ± 0.36	n.s
ALAT postop max (µkat/L)	0.57 ± 0.32	0.45 ± 0.28	p < 0.02
CKMB preop (µg/L)	4.01 ± 1.61	4.20 ± 2.29	n.s
CKMB postop max (µg/L)	27.69 ± 45.86	17.06 ± 20.70	n.s
CRP preop (mg/L)	8.1 ± 12.5	8.6 ± 12.4	n.s
CRP postop max (mg/L)	214.6 ± 65.6	215.7 ± 75.6	n.s
Leucocytes preop (10^9 /L)	7.73 ± 1.88	7.44 ± 1.64	n.s
Leucocytes postop max (10^9 /L)	12.56 ± 3.22	12.59 ± 3.25	n.s
Platelets preop (10^9 /L)	243.1 ± 78.1	239.9 ± 72.9	n.s
Platelets postop max (10^9 /L)	254.0 ± 74.9	272.2 ± 137.6	n.s
Cystatin C preop (mg/L)	1.14 ± 0.42	1.16 ± 0.38	n.s
Cystatin C postop max (mg/L)	1.42 ± 0.64	1.42 ± 0.60	n.s
Creatinine preop (µmol/L)	85.9 ± 27.8	86.7 ± 26.3	n.s
Creatinine postop max (µmol/L)	100.4 ± 48.3	102.8 ± 47.4	n.s
Urea preop (mmol/L)	7.39 ± 3.44	7.05 ± 2.51	n.s
Urea postop max (mmol/L)	9.07 ± 8.74	8.16 ± 4.26	n.s

Control group – group with the use of cardiotomy suction. Intended study group – group with the intention not to use cardiotomy suction. Values are expressed as mean ± 1 standard deviation or number (percent). Abbreviations: preop, preoperative; postop max, maximum value recorded postoperatively; n.s, no significant difference.

Table 14.
Peroperative and postoperative variables.

	Control group	Intended study group	p
n	71	75	
CPB time minutes	81.8 ± 25.1	74.0 ± 24.3	n.s
Aortic cross-clamping time minutes	49.5 ± 15.4	43.3 ± 15.3	p < 0.02
Number of grafts	2.61 ± 0.64	2.8 ± 0.6	n.s
Peroperative fluid balance ml	3582.9 ± 1186.8	3484.9 ± 1193.9	n.s
Peroperative diuresis ml	689.9 ± 399.2	607.1 ± 297.5	n.s
Peroperative norepinephrine µg	650 ± 723	510 ± 757	n.s
Postoperative norepinephrine µg	2100 ± 2890	1712 ± 1827	n.s [a]
Postoperative ventilator time hours	8.40 ± 9.60	9.45 ± 15.31	n.s [a]
Postoperative bleeding ml	850.4 ± 697.3	766 ± 536	n.s
Postoperative infektion no (%)	2 (2.8)	3 (4.0)	n.s
Postoperative inotropics >24 h no (%)	5 (7.0)	5 (6.7)	n.s
Postoperative ICU time hours	29.51 ± 22.66	26.15 ± 18.71	n.s [a]
Reoperation due to bleeding no (%)	1 (1.4)	2 (2.7)	n.s

Control group – group with the use of cardiotomy suction. Intended study group – group with the intention not to use cardiotomy suction, but where it had to be used because of blood loss. Values are expressed as mean ± 1 standard deviation or number (percent). [a] - comparison is made with Mann-Whitney U-test. Abbreviations: CPB, cardiopulmonary bypass; ICU, intensive care unit; n.s, no significant difference.

Table 15.
Baseline characteristics.

	Study group	X-over group	p
n	52	23	
Age	70.3 ± 9.7	68.8 ± 10.6	n.s
Female gender n (%)	7 (13.5)	7 (30.0)	n.s
Preoperative Euroscore	4.2 ± 2.4	5.4 ± 2.4	n.s
Diabetes n (%)	12 (23.1)	6 (26.1)	n.s
COPD n (%)	2 (3.8)	3 (13.0)	n.s
LVEF > 50% n (%)	38 (73.1)	12 (52.2)	n.s
LVEF 30-50% n (%)	12 (23.1)	9 (39.1)	n.s
LVEF < 30% n (%)	2 (3.8)	2 (8.7)	n.s
NYHA class			
I-II n (%)	36 (69.2)	13 (56.5)	n.s
III-IV n (%)	16 (30.8)	10 (43.5)	n.s
CCS class			
I-II n (%)	17 (32.7)	2 (8.7)	p < 0.05
III-IV n (%)	35 (67.3)	21 (91.3)	p < 0.05

Study group – group without the use cardiotomy suction. X-over group – group with the intention not to use cardiotomy suction, but where it had to be used because of blood loss. Values are expressed as mean ± 1 standard deviation or number (percent). Abbreviations: COPD, chronic obstructive pulmonary disease; LVEF, left ventricular ejection fraction; NYHA, New York Heart Association class; CCS, Canadian Cardiovascular Society class; n.s, no significant difference.

Table 16.
Transfusion of blood products and B-hemoglobin analysis.

	Study group	X-over group	p
n	52	23	
Hemoglobin preop (g/L)	139.0 ± 13.3	136.7 ± 13.6	n.s
Hemoglobin postop max (g/L)	106.8 ± 10.9	104.0 ± 7.9	n.s
Hemoglobin postop min (g/L)	91.9 ± 10.2	87.3 ± 9.4	n.s
Peroperative			
Received red blood cell transfusion n (%)	7 (13.5)	6 (26.1)	n.s
Transfused red blood cells units	0.19± 0.53	0.56 ± 1.34	n.s
Received plasma transfusion n (%)	1 (1.9)	2 (8.7)	n.s
Transfused plasma units	0.04 ± 0.28	0.26 ± 0.92	n.s
Received platelet transfusion n (%)	3 (5.8)	1 (4.4)	n.s
Transfused platelet units	0.10 ± 0.41	0.09 ± 0.42	n.s
Extended over the whole study period			
Received red blood cell transfusion n (%)	23 (44.2)	15 (65.2)	n.s
Transfused red blood cells units	1.12 ± 1.50	2.30 ± 2.38	$p < 0.02$
Received plasma transfusion n (%)	6 (11.5)	9 (39.1)	$p < 0.01$
Transfused plasma units	0.44 ± 1.54	2.04 ± 4.02	$p < 0.02$
Received platelet transfusion n (%)	3 (5.8)	6 (26.1)	$p < 0.02$
Transfused platelet units	0.10 ± 0.41	0.61 ± 1.23	$p < 0.01$

Study group – group without the use of cardiotomy suction. X-over group – group with the intention not to use cardiotomy suction, but where it had to be used because of blood loss. Values are expressed as mean ± 1 standard deviation or number (percent). Abbreviations: preop, preoperative; postop max, maximum value recorded postoperatively; postop min, minimum value recorded postoperatively; n.s, no significant difference.

Table 17.
Laboratory analysis.

	Study group	X-over group	p
n	52	23	
ASAT preop (µkat/L)	0.50 ± 0.19	0.50 ± 0.21	n.s
ASAT postop max (µkat/L)	0.63 ± 0.24	0.78 ± 0.46	n.s
ALAT preop (µkat/L)	0.57 ± 0.33	0.56 ± 0.43	n.s
ALAT postop max (µkat/L)	0.44 ± 0.20	0.49 ± 0.42	n.s
CKMB preop (µg/L)	4.10 ± 1.85	4.41 ± 3.10	n.s
CKMB postop max (µg/L)	15.82 ± 18.85	19.82 ± 24.58	n.s
CRP preop (mg/L)	8.9 ± 13.8	7.8 ± 8.3	n.s
CRP postop max (mg/L)	213.7 ± 73.5	220.1 ± 81.6	n.s
Leucocytes preop (10^9 /L)	7.47 ± 1.71	7.36 ± 1.50	n.s
Leucocytes postop max (10^9 /L)	13.0 ± 3.3	11.6 ± 2.9	n.s
Platelets preop (10^9 /L)	235.4 ± 69.3	250.0 ± 81.3	n.s
Platelets postop max (10^9 /L)	275.3 ± 151.3	265.2 ± 102.5	n.s
Cystatin C preop (mg/L)	1.18 ± 0.42	1.09 ± 0.26	n.s
Cystatin C postop max (mg/L)	1.46 ± 0.69	1.34 ± 0.34	n.s
Creatinine preop (µmol/L)	89.4 ± 29.0	80.7 ± 18.0	n.s
Creatinine postop max (µmol/L)	109.4 ± 53.8	87.8 ± 23.0	n.s
Urea preop (mmol/L)	7.30 ± 2.73	6.48 ± 1.84	n.s
Urea postop max (mmol/L)	8.64 ± 4.76	7.07 ± 2.64	n.s

Study group – group without the use of cardiotomy suction. X-over group – group with the intention not to use cardiotomy suction, but where it had to be used because of blood loss. Values are expressed as mean ± 1 standard deviation or number (percent). Abbreviations: preop, preoperative; postop max, maximum value recorded postoperatively; n.s, no significant difference.

Table 18.
Peroperative and postoperative variables.

	Study group	X-over group	p
n	52	23	
CPB time, minutes	69.5 ± 19.5	84.3 ± 30.8	p < 0.02
Aortic cross-clamping time minutes	42.8 ± 13.2	44.3 ± 19.7	n.s
Number of grafts	2.7 ± 0.6	2.8 ± 0.7	n.s
Peroperative fluid balance ml	3260.6 ± 879.0	3991.9 ± 1617.2	p < 0.02
Peroperative diuresis ml	534.6 ± 225.3	778.6 ± 375.0	p < 0.001
Peroperative norepinephrine µg	395 ± 344	769 ± 1245	p < 0.05
Postoperative norepinephrine µg	1648 ± 1678	1859 ± 2160	n.s [a]
Postoperative ventilator time hours	6.88 ± 3.08	15.16 ± 26.58	p < 0.05 [a]
Postoperative bleeding ml	702 ± 467	919 ± 666	n.s
Postoperative infektion no (%)	2 (3.8)	1 (4.4)	n.s
Postoperative inotropics >24 h no (%)	2 (3.8)	3 (13.0)	n.s
Postoperative ICU time hours	23.30 ± 8.36	32.46 ± 30.70	n.s [a]
Reoperation due to bleeding no (%)	2 (3.8)	0 (0.0)	n.s

Study group – group without the use of cardiotomy suction. X-over group – group with the intention not to use cardiotomy suction, but where it had to be used because of blood loss. Values are expressed as mean ± 1 standard deviation or number (percent). [a] - comparison is made with Mann-Whitney U-test. Abbreviations: CPB, cardiopulmonary bypass; ICU, intensive care unit; n.s, no significant difference.

Discussion

This thesis is based on four studies. Three of them (I, II and IV) mainly addressed questions concerning cardiac surgery and cardiotomy suction. Two of the studies (I and II) were experimental studies introducing a novel porcine model. Undertaking these studies, we have been able to demonstrate the differential distribution of LME in cardiac surgery (study I), and examine the kinetics of lipid microembolism in general and the renal kinetics in particular during cardiac surgery (study II).

The two remaining studies were clinical studies on patients scheduled for CABG. In study III, we followed the findings from study I and II regarding the kidney. It is well known that renal dysfunction is a problem in cardiac surgery [18]. Cystatin C is known to be a better marker for GFR than creatinine [28, 31, 32, 51, 52], which is the most common clinically used marker of GFR to day [28]. We wanted to evaluate cystatin C in cardiac surgery, and hopefully find a better tool for monitoring renal function postoperatively. In the final study (IV), we examined common outcome variables in cardiac surgery, comparing surgery without cardiotomy suction with surgery including cardiotomy suction, the later with an obviously increased risk of introducing lipid particles into the blood circulation.

The organ that has been of focus in previous studies on LME has primarily been the brain.[5, 6, 10, 43]. In study I, we found that lipid particles introduced into the cardiopulmonary circuit, not only affected the brain, but was a global phenomenon affecting all organs. In addition, the variation from organ to organ was large, with the kidney, liver, and spleen seeming to have the highest embolic load.

The kidney was found to be the organ with the highest level of beta radiation, and therefore believed to have the highest uptake of LME. Since the kidney is a well perfused organ [64], it was to be expected that it would receive a relatively large load of LME. However, the extremely high levels found in our study were surprising. In a study by Boston and colleagues [65] blood flow to different organs was studied in a porcine model. They found a higher blood flow to the brain than to the kidney. In our study, we found nearly a 10-fold higher beta radiation level in the kidney than in the brain. This finding indicates that

it is not only the blood flow to the kidney that is responsible for the uptake of LME.

As mentioned earlier, renal dysfunction is a well known complication after cardiac surgery [18]. Since study I clearly showed that the kidney had the greatest uptake of LME, a relevant question is whether LME are a contributing factor to this complication. At least two different potential mechanisms responsible for organ dysfunction by lipid emboli are possible. Mechanical obstruction is of course one of them. It has been shown in vitro that mediastinal fat clogs filters and impairs circulation [66]. Our study showed a high uptake of lipid material in the kidney, and it is therefore likely that these emboli could cause a mechanical obstruction to blood flow, which could contribute to an impaired renal function. Another explanation could be a toxic reaction. It has been shown that oleic acid, when given intravenously to test animals, causes lung injury with edema and severe hypoxemia [67]. In addition, it has been shown that uncharged fat (such as triglycerides) and also free fatty acids have toxic properties. In a feline model, triolein and oleic acid cause both vasogenic and cytotoxic cerebral edema, when they were infused into the carotid artery [60]. In that study, the charged oleic acid caused the greatest damage. This implies that lipid material can not only cause mechanical obstruction but chemical interactions may also play a negative role in the capillaries of the organs.

At first glance, the overall level of beta radiation in the brain was lower than anticipated. Compared with the kidney and the spleen for example, the brain level was only a fraction of these levels, and could therefore be assumed to be of less importance. However, the porcine model used may not be completely representative of the clinical setting. Cerebral blood flow in pigs is somewhat lower than in humans [64, 65, 68, 69]. Pigs also have a rete mirabile, which could affect the rate of embolization. This is a vascular rete situated between the carotid arteries and cerebral vessels, which acts as a thermoregulator [70]. The small vessels of the rete could work as a screen filter for emboli. Our study corroborates previous findings that LME from shed mediastinal blood causes massive microembolization in the brain [6, 10]. The question of the relation between these emboli and the cognitive dysfunction seen after surgery is not addressed in our study, but the connection between the two entities is intriguing and have been suggested [17, 22, 71]. Our study did, however, reveal differences between different regions of the brain (Table 2). One interesting observation was that we found the highest levels of beta radiation in the gray matter of the cerebrum, which is involved in cognitive functions of the brain [72]. This finding will further spur the debate on the possible relation between LME and postoperative cognitive dysfunction.

In study II, we demonstrated the rapid kinetics of LME during cardiac surgery with CPB in the systemic circulation. Twenty seconds after infusion, LME reached the arterial system, and after a median of 40 seconds peak levels

were found in the blood. These emboli were eliminated quickly as seen in Figure 2. A certain degree of recirculation seemed to occur, since a second peak was found in all but one animal, and in some animals even a third peak. During the first 20 minutes of the experiment, about 2/5 of the radioactivity found in the arterial blood was also found in the venous blood. 3/5 seemed to be trapped in the capillary system, indicating that the capillary system acts as a highly efficient filter trapping these emboli.

In the renal group, we specifically studied the elimination of LME through the kidney. The difference in the 15 minutes AUC between the renal artery and the renal vein showed that only approximately 23% of the LME´s passed the kidneys, meaning that 77% of the LME´s were trapped inside the kidneys. This part of the study was made on only four animals that did not undergo CPB. Therefore, we have to be very cautious in the interpretation of these finding. However, it seems as if the kidney trap more particles in the first passage than the body as a whole, which could to some part explain the higher levels of lipid particles in the kidney as compared to other organs.

The study also shed some light on the metabolism of these lipid particles. In the systemic group virtually no radioactivity was found in the urine of the animals when sacrificed (approximately 30 minutes after the infusion of shed blood). This finding suggests that no measurable renal excretion takes place during this time period.

The infusion of shed blood in the systemic group was followed by a transient decline in blood pressure. Since the animals were on CPB, even though the aorta was not X-clamped, the majority of the flow was controlled by the heart-lung machine. As the pump flow was kept constant, any decrease in blood pressure would probably be due to changes in peripheral resistance and not flow. Another study by Westerberg and associates [73] have shown that cardiotomy suction blood, when infused quickly as a bolus into the by-pass circuit during coronary artery bypass grafting, can lead to a similar transient fall in systemic blood pressure. It was argued that cardiotomy suction blood reduces vascular resistance due to an inflammatory response in the vasculature. Our study offers no explanation for the finding of a decrease in blood pressure.

Even though all animals received shed blood from a shed blood phantom with equal amount of radiation, the amount of radiation measured in the animals differed from animal to animal. The relative radiation measured between different organs did not differ much from animal to animal (Figure 1). We made an effort to blend the content of the shed blood phantom before retransfusing the shed blood into the CPB circuit. Still, fat and blood do not mix well, which is illustrated in Figure 6. The rubber tubing and the oxygenator in the CPB circuit are lipophilic and an educated guess is that lipids in the blood probably were trapped there as well as in the different organs. A recent submitted, but yet unpublished, study by Eyjolfsson and associates [74] showed that if the lipids

were emulsified (and therefore more water soluble) before being retransfused into the CPB circuit in a similar porcine study as in study I, it led to a higher and a more even uptake off LME between the test animals.

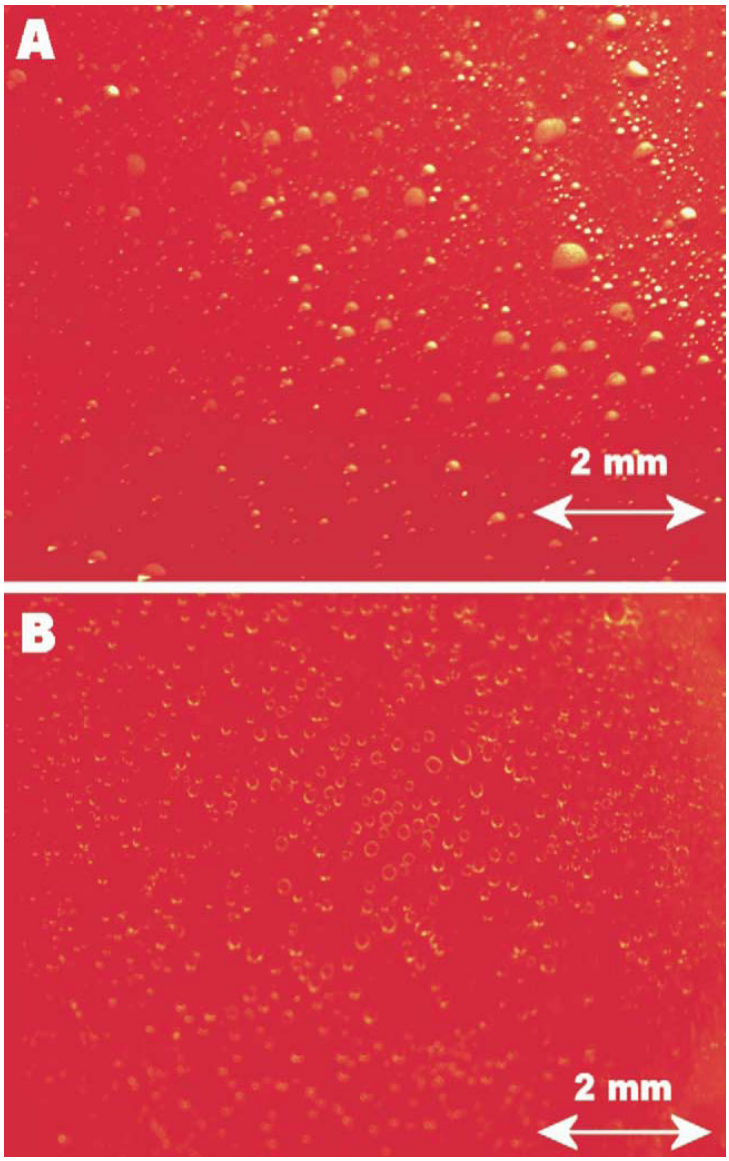

Figure 6.

(A) Macrophotograph of human shed blood from the surgical field containing triglycerides. (B) Macrophotograph of porcine blood with radioactive triglyceride added.

Creatinine is the most commonly used marker of GFR and renal function [28, 29], but cystatin C has been reported to be superior to creatinine in assessing renal function [30-32, 34]. In study III, we evaluated P-cystatin C with iohexol clearance (a reference method for determining GFR [41, 42]), used for the first time in cardiac surgery. The study revealed a strong correlation between postoperative iohexol clearance and postoperative cystatin C (Table 6). This corroborated well with other studies made on surgical as well as nonsurgical patients where cystatin C has been compared with a GFR reference method [30, 32, 39, 41]. A subgroup of patients with a postoperative iohexol clearance < 90 ml/min/1.73m^2 was formed. The group was created because it represents patients where monitoring of renal function is important, excluding patients with normal renal function. This subgroup showed an even stronger correlation between postoperative iohexol clearance and P-cystatin C compared with the entire study group (Table 6). In this group, the strength of the correlation between estimated cystatin C clearance (eCyC) and iohexol clearance vs. estimated creatinine clearance (CG and MDRD) and iohexol clearance were comparable (Table 6). However, cystatin C-based estimations seem superior in approximating actual GFR as no difference was found compared with iohexol clearance (Table 7). In contrast, creatinine-based clearance estimations yielded an overestimation of 30–40%. Therefore, the relative superiority of cystatin C seems greater in patients with a decreased GFR, where accurate monitoring of renal function is important.

Even though it has not been suggested that cystatin C is an acute-phase protein, earlier studies have shown a correlation to CRP [75, 76] and logarithmic CRP concentrations [77]. As the inflammatory response is present in all surgical patients to a varying degree, this could be relevant. However, we did not find any postoperative correlation between P-cystatin C and P-CRP or logarithmic P-CRP concentrations, alone or in multivariate analysis with iohexol clearance. Our findings are in concordance with a recent study by Grubb and associates [78] where they state that cystatin C is not influenced by inflammation. Therefore, it is likely that P-cystatin C in our study specifically reflects renal function and not inflammatory response.

The most accurate method to determine GFR is to inject and determine the renal elimination of an exogenous substance such as inulin, ^{51Cr}EDTA or iohexol [42]. However, these techniques are complicated, time-consuming, costly and have potential side effects. They are therefore not suitable for day to day surveillance of renal function in clinical settings. We chose the iohexol technique because it is easy to handle, well documented and does not involve radiation. However, high doses of iohexol can be nephrotoxic. In our study, the patients received a small amount of iohexol pre- and postoperatively (just a fraction of the dose given duringa normal coronary angiography) and the risk should therefore be negligible [79].

The final paper (IV) describes the clinical study we launched including 150 patients scheduled for CABG. The time span for completing the study was three years from the enrolment of the first patient to the gathering of the last data.

The study did not reveal any considerable disadvantages in outcome when CABG was performed without cardiotomy suction. The few differences revealed in this study, actually favored CABG without the use of cardiotomy suction.

The use of cardiotomy suction was introduced during the sixties. The aim was to reduce blood loss and the need of blood transfusion [9]. Blood transfusion in cardiac surgery has been associated with increased long-term mortality [80-82]. Our study could not reveal an increased rate of transfusion of blood products in the study group compared with the control group. On the contrary, in this study population, the control group received significantly more transfused units of packed red blood cells during surgery, as well as throughout the whole study period as compared with the study group (per protocol analysis)(Table 9). The study also revealed that a significantly larger percentage of patients in the control group received plasma transfusion, and if the X-over group was added to the control group, also the amount of transfused units of plasma became significantly larger compared with the study group (Table 9). If one compared the intended study group with the control group regarding transfusion of blood products, it tended to be an over all higher transfusion in the control group with the exception of platelet transfusion, which tended to be higher in the intention to treat group (Table 12). The differences were not significant. If the study group and the X-over group were compared, transfusion of blood products was significantly higher in the X-over group extended over the whole study period, but not if one only examined the peroperative transfusion rate (Table 16). The intended study group was of course a mixed group, where some patients had surgery with cardiotomy suction and some without. The important thing, that can be concluded, was that the outcome regarding transfusion of blood products (or other outcome variables) did not swing to and fro depending on whether the X-over group was included or not (see results).

Recent studies have suggested adverse effects of cardiotomy suction [2-6, 10, 13-15, 17, 73] and studies have been performed where cardiotomy suction was not used and collected shed blood was washed with a cell-saver [83, 84]. These studies indicated an increased blood product usage and increased postoperative bleeding when cell-savers were used as compared with normal cardiotomy suction. Our study does not suggest that cardiotomy suction is necessarily beneficial for reducing the need of transfusion of blood products in this selected group of patients. The study has shown that it is possible to conduct CABG with CPB with less transfusion of blood products, without any salvage of shed blood or retransfusion of shed blood in any form. To achieve these results, we believe it is important that the surgeon from the start of the operation is careful in creating a surgical field with as little unnecessary bleeding as possible,

instead of relying on an indiscriminate use of cardiotomy suction in a routine manner. The cardiotomy suction device must, of course, be ready for immediate use during the operation, if it becomes inevitable to refrain from using it.

Postoperatively, P-ASAT and P-ALAT were significantly higher in the control group (Table 10). The most suitable theory for this finding is that the manipulation of the heart during surgery and local tissue injury excrete enzymes into the wound cavity, which are retransfused to the patient by the cardiotomy suction [85, 86]. An alternative explanation could be that the control group had a significant longer aortic clamping time and CPB time compared to the study group (Table 11). This would theoretically produce more ischemia in the myocardium and could cause the elevation of the enzymes postoperatively. A combination of both could also explain this finding.

Previous studies have reported a decline in systemic vascular resistance due to vasodilatation and a consequent fall in blood pressure, as a result of the use of cardiotomy suction [73, 87]. In the present study, where norepinephrine was primarily used for the treatment of vasodilatation, a peroperative increased usage of norepinephrine was shown in the control group compared with the study group (Table 11). This finding could indicate that there was an increased vasodilatation in the control group compared with the study group. Crystalloids were mainly used for volume expansion during surgery, and blood products were only used, when the physician in charge found it necessary. Peroperative diuresis was significantly higher in the control group (Table 11) and the fluid balance during surgery had a tendency of being larger in the control group compared with the study group (Table 11). If the 23 X-over cases were added to the control group the difference became significant (Table 11). The tendency for higher fluid balance in the control group was in line with the higher need for norepinephrine in the same group mentioned above, since fluid infusion obviously can compensate for vasodilatation.

There were no differences in P-cystatin C, P-creatinine or P-urea between the two groups preoperatively or postoperatively (Table 10). With the knowledge we gained in study I and II concerning the large renal deposit of LME, one could have expected to find that the present study (IV) should have revealed a difference between the two groups in at least P-cystatin C. If there was a difference in kidney function between the groups it was not detectable through available laboratory analysis. However, surgeons probably try to minimize the bleeding during surgery, irrespectively of the usage of cardiotomy suction or not. As a favorable consequence, the embolic load tends to become small in routine CABG patients even though cardiotomy suction is used. Therefore, in operation with larger blood loss a detectable impairment of renal function could potentially be seen.

We have mainly focused on the few differences that were found between the groups in study IV, but we should not forget that most of the investigated

outcome variables did not differ between the control group and the study group. All in all, we could conclude that CABG with CPB can be conducted safely without using cardiotomy suction and retransfusion of shed blood as long as the surgeon is vigilant on the blood loss, and prepared to use cardiotomy suction when needed.

Conclusions

Study I
This study showed that embolization of lipids is not a phenomenon restricted to the brain, but affected all the organs examined. The kidney was the organ that trapped most LME. The grey matter of the brain was the part, within the brain, that received most LME.

Study II
This study showed that lipid emboli infused by means of cardiotomy suction will quickly reach the circulation of the patient. There was a high degree of first-pass trapping in the capillaries. No obvious immediate renal excretion of lipid material was found.

Study III
This study indicated that clearance estimations based on plasma cystatin C are superior in accuracy compared with estimations based on plasma creatinine in determining GFR in cardiac surgery. Plasma cystatin C had, in this study population, a stronger correlation to iohexol clearance compared with the correlation between plasma creatinine and iohexol clearance. Plasma cystatin C can be used accurately as a marker of GFR and renal function postoperatively in cardiac surgery.

Study IV
This study showed that CABG with CPB can be conducted safely without using cardiotomy suction and retransfusion of shed blood, as long as the surgeon is vigilant on the blood loss and prepared to use cardiotomy suction when needed.

Grants

The following grants have funded this thesis:

Swedish Heart-Lung Foundation
Crafoord Foundation, Lund, Sweden.
Swedish governmental grants (ALF – "Avtalet om Läkarutbildning och Forskning").
Region Skåne research funds
Lund University Hospital funds

Acknowledgements

Henrik Bjursten, i mina ögon har du förkroppsligat essensen av vad gott handledararskap står för. Du har varit vägledande, undervisande, stöttande, respektfull och fram för allt närvarande. Med dig som handledare har forskning blivit en källa till personlig glädje och utveckling för mig.

Sten Blomquist, för att jag tidigt skulle kunna bli doktorandanmäld tog du initialt rollen som huvudhandledare och trädde osjälviskt tillbaks, när Henrik var redo att formellt ta befälet. Jag har haft många roliga diskussioner med dig, som inte enbart rört sig om forskning. Du har dessutom varit min ledstjärna genom åren när det gällt thoraxanestesiologi.

Magnus Dencker, utan din hjälp och kunskap hade området kring radioaktivitet och scintillation blivit ett rent mörker. Tack vare dig blev det ju istället alldeles strålande.

Mats Allers, förutom att du förtjänstfullt skötte perfusionen under den laborativa delen till denna avhandling, gjorde du ditt yttersta för att höja det allmänna stämningsläget under våra försök.

Atli Eyjofsson, min gode doktorandkompis, som rent praktiskt har varit involverad i en betydande del av materialet till den kliniska delen av avhandlingen.

Per Ederoth, mitt första minne av dig är när du var Toddyfar och jag Toddygubbe under Toddydagen min första termin i Lund. Klarare minnen har jag från utmanande och spännande off pist åkning, där du ledde vägen. Du har haft många goda synpunkter som hjälpt mig i mitt skrivande. Dessutom har vi haft en del diskussioner kring ”Formel 1” som varit upplyftande.

Alain Dardashti, min gode doktorankompis, med vem jag delar ett ”spinnande” intresse.

Ignacio Plaza, för att du med stor energi och glädje alltid ställde upp på labbet.

Peter Höglund, för goda råd kring frågor gällande statistik.

Sten-Erik Bäck, jag vill tacka dig för din stora hjälp kring frågor gällande iohexol clearance.

Bo Frenby, för goda råd kring clearance beräkningar.

Anders Grubb, för goda råd i samband med planeringen av de kliniska studierna.

Bertil Persson och **Gustav Grafstöm**, för er hjälp med isotoper.

Lars-Erik Nilsson, för din tekniska assistans och ditt tillhandahållande av programvara för scintillationsräkning.

Kollegorna inom thoraxanestesi och thoraxkirurgi, för uppmuntran och moralisk stöttning.

Personalen på thorax anestesi, THIVA samt avd 17, 18 och 19, för hjälp med provtagning och datainsamling.

Lars Algotsson, du hade en avgörande betydelse för att jag valde att specialisera mig till anestesiolog, vilket jag inte ångrat.

Birgitta, Bengt och **Rosalie,** för allmänt stöd.

Wilmer och Hampus, ni är mina spinnande, mysiga och luddiga grabbar som tillför vårt hem stor trevnad.

Johan och Elin, mina bonusbarn, som ger mig så mycket glädje i vardagen.

Frida och Karolina, mina älskade döttrar. Det är ni som är det viktigaste för mig.

Karin, mitt livs kärlek. Jag blir glad så fort jag ser dig.

References

1. Edmunds, L.H., ed. *Cardiac surgery in the adult*. 1997: p. 269-270.
2. Skrabal, C.A., et al., *Pericardial suction blood separation attenuates inflammatory response and hemolysis after cardiopulmonary bypass.* Scand Cardiovasc J, 2006. **40**(4): p. 219-23.
3. Svitek, V., V. Lonsky, and F. Anjum, *Pathophysiological aspects of cardiotomy suction usage.* Perfusion. **25**(3): p. 147-52.
4. Westerberg, M., A. Bengtsson, and A. Jeppsson, *Coronary surgery without cardiotomy suction and autotransfusion reduces the postoperative systemic inflammatory response.* Ann Thorac Surg, 2004. **78**(1): p. 54-9.
5. Brooker, R.F., et al., *Cardiotomy suction: a major source of brain lipid emboli during cardiopulmonary bypass.* Ann Thorac Surg, 1998. **65**(6): p. 1651-5.
6. Brown, W.R., D.M. Moody, and V.R. Challa, *Cerebral fat embolism from cardiopulmonary bypass.* J Neuropathol Exp Neurol, 1999. **58**(2): p. 109-19.
7. Eyjolfsson, A., et al., *Characterization of lipid particles in shed mediastinal blood.* Ann Thorac Surg, 2008. **85**(3): p. 978-81.
8. Jonsson, H., et al., *Circulating particles during cardiac surgery.* Interact Cardiovasc Thorac Surg, 2009. **8**(5): p. 538-42.
9. Lau, K., et al., *Coronary artery surgery: cardiotomy suction or cell salvage?* J Cardiothorac Surg, 2007. **2**: p. 46.
10. Moody, D.M., et al., *Brain microemboli associated with cardiopulmonary bypass: a histologic and magnetic resonance imaging study.* Ann Thorac Surg, 1995. **59**(5): p. 1304-7.
11. Appelblad, M. and K.G. Engstrom, *Fat content in pericardial suction blood and the efficacy of spontaneous density separation and surface adsorption in a prototype system for fat reduction.* J Thorac Cardiovasc Surg, 2007. **134**(2): p. 366-72.
12. Kincaid, E.H., et al., *Processing scavenged blood with a cell saver reduces cerebral lipid microembolization.* Ann Thorac Surg, 2000. **70**(4): p. 1296-300.

13. Svenmarker, S. and K.G. Engstrom, *The inflammatory response to recycled pericardial suction blood and the influence of cell-saving.* Scand Cardiovasc J, 2003. **37**(3): p. 158-64.
14. Jonsson, H., *The rationale for intraoperative blood salvage in cardiac surgery.* J Cardiothorac Vasc Anesth, 2009. **23**(3): p. 394-400.
15. Nakahira, A., et al., *Cardiotomy suction, but not open venous reservoirs, activates coagulofibrinolysis in coronary artery surgery.* J Thorac Cardiovasc Surg.
16. de Haan, J., et al., *Retransfusion of suctioned blood during cardiopulmonary bypass impairs hemostasis.* Ann Thorac Surg, 1995. **59**(4): p. 901-7.
17. Taggart, D.P. and S. Westaby, *Neurological and cognitive disorders after coronary artery bypass grafting.* Curr Opin Cardiol, 2001. **16**(5): p. 271-6.
18. Mangano, C.M., et al., *Renal dysfunction after myocardial revascularization: risk factors, adverse outcomes, and hospital resource utilization. The Multicenter Study of Perioperative Ischemia Research Group.* Ann Intern Med, 1998. **128**(3): p. 194-203.
19. Gardner, T.J., et al., *Stroke following coronary artery bypass grafting: a ten-year study.* Ann Thorac Surg, 1985. **40**(6): p. 574-81.
20. Mickleborough, L.L., et al., *Risk factors for stroke in patients undergoing coronary artery bypass grafting.* J Thorac Cardiovasc Surg, 1996. **112**(5): p. 1250-8; discussion 1258-9.
21. Benedict, R.H., *Cognitive function after open-heart surgery: are postoperative neuropsychological deficits caused by cardiopulmonary bypass?* Neuropsychol Rev, 1994. **4**(3): p. 223-55.
22. de Vries, A.J., et al., *Clinical evaluation of a new fat removal filter during cardiac surgery.* Eur J Cardiothorac Surg, 2004. **25**(2): p. 261-6.
23. Stafford-Smith, M., *Evidence-based renal protection in cardiac surgery.* Semin Cardiothorac Vasc Anesth, 2005. **9**(1): p. 65-76.
24. Brown, J.R., et al., *Long-term survival after cardiac surgery is predicted by estimated glomerular filtration rate.* Ann Thorac Surg, 2008. **86**(1): p. 4-11.
25. Hobson, C.E., et al., *Acute kidney injury is associated with increased long-term mortality after cardiothoracic surgery.* Circulation, 2009. **119**(18): p. 2444-53.
26. Holzmann, M.J., et al., *Renal insufficiency and long-term mortality and incidence of myocardial infarction in patients undergoing coronary artery bypass grafting.* Eur Heart J, 2007. **28**(7): p. 865-71.
27. Bihorac, A., et al., *Long-term risk of mortality and acute kidney injury during hospitalization after major surgery.* Ann Surg, 2009. **249**(5): p. 851-8.
28. Johnson, D., *The CARI guidelines. Evaluation of renal function.* Nephrology (Carlton), 2005. **10 Suppl 4**: p. S133-76.

29. Mathew, T.H., *Chronic kidney disease and automatic reporting of estimated glomerular filtration rate: a position statement.* Med J Aust, 2005. **183**(3): p. 138-41.
30. Dharnidharka, V.R., C. Kwon, and G. Stevens, *Serum cystatin C is superior to serum creatinine as a marker of kidney function: a meta-analysis.* Am J Kidney Dis, 2002. **40**(2): p. 221-6.
31. Filler, G., et al., *Cystatin C as a marker of GFR--history, indications, and future research.* Clin Biochem, 2005. **38**(1): p. 1-8.
32. Fliser, D. and E. Ritz, *Serum cystatin C concentration as a marker of renal dysfunction in the elderly.* Am J Kidney Dis, 2001. **37**(1): p. 79-83.
33. Nergelius, G., et al., *Renal dysfunction after total knee replacement is not aggravated by bone cement.* Acta Anaesthesiol Scand, 1998. **42**(8): p. 974-81.
34. Newman, D.J., et al., *Serum cystatin C measured by automated immunoassay: a more sensitive marker of changes in GFR than serum creatinine.* Kidney Int, 1995. **47**(1): p. 312-8.
35. Abu-Omar, Y., et al., *Evaluation of Cystatin C as a marker of renal injury following on-pump and off-pump coronary surgery.* Eur J Cardiothorac Surg, 2005. **27**(5): p. 893-8.
36. Haase, M., et al., *Novel biomarkers early predict the severity of acute kidney injury after cardiac surgery in adults.* Ann Thorac Surg, 2009. **88**(1): p. 124-30.
37. Haase-Fielitz, A., et al., *Novel and conventional serum biomarkers predicting acute kidney injury in adult cardiac surgery--a prospective cohort study.* Crit Care Med, 2009. **37**(2): p. 553-60.
38. Heise, D., et al., *Utility of cystatin C for assessment of renal function after cardiac surgery.* Nephron Clin Pract, 2009. **112**(2): p. c107-14.
39. Qing-Ping W, J.-W.G., Xiang-Hong Z, Hui L, Xiang-Hang L, *Assessment of glomerular filtration rate by serum cystatin C in patients undergoing coronary artery bypass grafting.* Ann Clin Biochem, 2009. **46**: p. 495-500.
40. Ristikankare, A., et al., *Serum cystatin C in elderly cardiac surgery patients.* Ann Thorac Surg, 2010. **89**(3): p. 689-94.
41. Kyhse-Andersen, J., et al., *Serum cystatin C, determined by a rapid, automated particle-enhanced turbidimetric method, is a better marker than serum creatinine for glomerular filtration rate.* Clin Chem, 1994. **40**(10): p. 1921-6.
42. Larsson, A., et al., *Calculation of glomerular filtration rate expressed in mL/min from plasma cystatin C values in mg/L.* Scand J Clin Lab Invest, 2004. **64**(1): p. 25-30.
43. Moody, D.M., et al., *Brain microemboli during cardiac surgery or aortography.* Ann Neurol, 1990. **28**(4): p. 477-86.

44. Carlsson, S., *Vätskescintillatorn.* Report LiH-RAD-R-021, Institution of Radiophysics, Lund University, 1975.
45. Olsson, B., et al., *Human pharmacokinetics of iohexol. A new nonionic contrast medium.* Invest Radiol, 1983. **18**(2): p. 177-82.
46. Gaspari, F., et al., *Plasma clearance of nonradioactive iohexol as a measure of glomerular filtration rate.* J Am Soc Nephrol, 1995. **6**(2): p. 257-63.
47. Back, S.E., E. Krutzen, and P. Nilsson-Ehle, *Contrast media as markers for glomerular filtration: a pharmacokinetic comparison of four agents.* Scand J Clin Lab Invest, 1988. **48**(3): p. 247-53.
48. Krutzen, E., et al., *Plasma clearance of a new contrast agent, iohexol: a method for the assessment of glomerular filtration rate.* J Lab Clin Med, 1984. **104**(6): p. 955-61.
49. Peters, A.M., *The kinetic basis of glomerular filtration rate measurement and new concepts of indexation to body size.* Eur J Nucl Med Mol Imaging, 2004. **31**(1): p. 137-49.
50. Sterner, G., et al., *Validation of a new plasma cystatin C-based formula and the Modification of Diet in Renal Disease creatinine-based formula for determination of glomerular filtration rate.* Scand J Urol Nephrol, 2009. **43**(3): p. 242-9.
51. Nilsson-Ehle, P. and A. Grubb, *New markers for the determination of GFR: iohexol clearance and cystatin C serum concentration.* Kidney Int Suppl, 1994. **47**: p. S17-9.
52. O'Riordan, S.E., et al., *Cystatin C improves the detection of mild renal dysfunction in older patients.* Ann Clin Biochem, 2003. **40**(Pt 6): p. 648-55.
53. Bokenkamp, A., et al., *Effect of corticosteroid therapy on serum cystatin C and beta2-microglobulin concentrations.* Clin Chem, 2002. **48**(7): p. 1123-6.
54. Fricker, M., et al., *Impact of thyroid dysfunction on serum cystatin C.* Kidney Int, 2003. **63**(5): p. 1944-7.
55. Cockcroft, D.W. and M.H. Gault, *Prediction of creatinine clearance from serum creatinine.* Nephron, 1976. **16**(1): p. 31-41.
56. Levey, A.S., et al., *A simplified equation to predict glomerular filtration rate from serum creatinine (abstract).* J Am Soc Nephrol, 2000 p. 11:115 A.
57. Grubb, A., et al., *Simple cystatin C-based prediction equations for glomerular filtration rate compared with the modification of diet in renal disease prediction equation for adults and the Schwartz and the Counahan-Barratt prediction equations for children.* Clin Chem, 2005. **51**(8): p. 1420-31.
58. Drew, P.A., E. Smith, and P.D. Thomas, *Fat distribution and changes in the blood brain barrier in a rat model of cerebral arterial fat embolism.* J Neurol Sci, 1998. **156**(2): p. 138-43.
59. Jacobovitz-Derks, D. and C.M. Derks, *Pulmonary neutral fat embolism in dogs.* Am J Pathol, 1979. **95**(1): p. 29-42.

60. Kim, H.J., et al., *Experimental cerebral fat embolism: embolic effects of triolein and oleic acid depicted by MR imaging and electron microscopy.* AJNR Am J Neuroradiol, 2002. **23**(9): p. 1516-23.
61. Nakata, Y., et al., *Triolein-induced pulmonary embolization and increased microvascular permeability in isolated perfused rat lungs.* J Trauma, 1999. **47**(1): p. 111-9.
62. Insull, W.j. and G. Bartsch, *Fatty acid composition of human adipose tissue related to age, sex and race.* Am J Clin Nutr, 1967. **20**: p. 13-23.
63. Brochner-Mortensen, J., *A simple method for the determination of glomerular filtration rate.* Scand J Clin Lab Invest, 1972. **30**(3): p. 271-4.
64. Selwyn, A.P., et al., *Regional myocardial and organ blood flow after myocardial infarction: application of the microsphere principle in man.* Circulation, 1986. **73**(3): p. 433-43.
65. Boston, U.S., et al., *Hierarchy of regional oxygen delivery during cardiopulmonary bypass.* Ann Thorac Surg, 2001. **71**(1): p. 260-4.
66. Appelblad, M. and G. Engstrom, *Fat contamination of pericardial suction blood and its influence on in vitro capillary-pore flow properties in patients undergoing routine coronary artery bypass grafting.* J Thorac Cardiovasc Surg, 2002. **124**(2): p. 377-86.
67. Julien, M., J.M. Hoeffel, and M.R. Flick, *Oleic acid lung injury in sheep.* J Appl Physiol, 1986. **60**(2): p. 433-40.
68. Floyd, T.F., et al., *Perioperative changes in cerebral blood flow after cardiac surgery: influence of anemia and aging.* Ann Thorac Surg, 2003. **76**(6): p. 2037-42.
69. Slater, J.M., T.A. Orszulak, and D.J. Cook, *Distribution and hierarchy of regional blood flow during hypothermic cardiopulmonary bypass.* Ann Thorac Surg, 2001. **72**(2): p. 542-7.
70. Burbridge, B., G. Matte, and A. Remedios, *Complex intracranial arterial anatomy in swine is unsuitable for cerebral infarction projects.* Can Assoc Radiol J, 2004. **55**(5): p. 326-9.
71. Stump, D.A., et al., *Emboli and Neuropsychological Outcome Following Cardiopulmonary Bypass.* Echocardiography, 1996. **13**(5): p. 555-558.
72. Kolb, B. and R. Tees, *The cerebral cortex of the rat.* Cambridge, MA: MIT Press, 1990.
73. Westerberg, M., et al., *Hemodynamic effects of cardiotomy suction blood.* J Thorac Cardiovasc Surg, 2006. **131**(6): p. 1352-7.
74. Eyjolfsson, A., et al., *Differences between emulsified and non-emulsified lipid micro embolization in an experimental model of mediastinal shed blood reinfusion.* (Submitted manuscript)
75. Keller, C.R., et al., *Kidney function and markers of inflammation in elderly persons without chronic kidney disease: the health, aging, and body composition study.* Kidney Int, 2007. **71**(3): p. 239-44.

76. Shlipak, M.G., et al., *Cystatin-C and inflammatory markers in the ambulatory elderly.* Am J Med, 2005. **118**(12): p. 1416.
77. Samouilidou, E.C. and E. Grapsa, *Relationship of serum cystatin C with C-reactive protein and apolipoprotein A1 in patients on hemodialysis.* Ren Fail, 2008. **30**(7): p. 711-5.
78. Grubb, A., et al., *Cystatin C, a marker for successful aging and glomerular filtration rate, is not influenced by inflammation.* Scand J Clin Lab Invest.
79. James, S., *Year report 2007.* Swedish Coronary Angiography and Angioplasty Registry (SCAAR), 2008: p. 100.
80. Engoren, M.C., et al., *Effect of blood transfusion on long-term survival after cardiac operation.* Ann Thorac Surg, 2002. **74**(4): p. 1180-6.
81. Koch, C.G., et al., *Transfusion in coronary artery bypass grafting is associated with reduced long-term survival.* Ann Thorac Surg, 2006. **81**(5): p. 1650-7.
82. Koch, C.G., et al., *Morbidity and mortality risk associated with red blood cell and blood-component transfusion in isolated coronary artery bypass grafting.* Crit Care Med, 2006. **34**(6): p. 1608-16.
83. Djaiani, G., et al., *Continuous-flow cell saver reduces cognitive decline in elderly patients after coronary bypass surgery.* Circulation, 2007. **116**(17): p. 1888-95.
84. Rubens, F.D., et al., *The cardiotomy trial: a randomized, double-blind study to assess the effect of processing of shed blood during cardiopulmonary bypass on transfusion and neurocognitive function.* Circulation, 2007. **116**(11 Suppl): p. I89-97.
85. De Paulis, R., et al., *Levels of troponin I and cardiac enzymes after reinfusion of shed blood in coronary operations.* Ann Thorac Surg, 1998. **65**(6): p. 1617-20.
86. Schmidt, H., et al., *Cardiac enzymes and autotransfusion of shed mediastinal blood after myocardial revascularization.* Ann Thorac Surg, 1997. **63**(5): p. 1288-92.
87. Eyjolfsson, A., et al., *Cardiorespiratory effects of venous lipid micro embolization in an experimental model of mediastinal shed blood reinfusion.* J Cardiothorac Surg, 2009. **4**: p. 48.
88. Rowland, M., ed. *Clinical Pharmacokinetics: Concepts and Applications.* 1980: p. 289.
89. DuBois, D. and E.F. DuBois, *A formula to estimate the approximate surface area if height and weight be known.* Arch Int Med, 1916. **17**: p. 863.