Summary

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In the past few decades ultrasound has developed to a widely available, easy to use diagnostic tool in routine clinical care. The introduction of microbubbles extended its diagnostic value. Not long thereafter, the therapeutic potential of contrast ultrasound on thrombus dissolution was investigated.

The aim of the work presented in this thesis was to evaluate possible new diagnostic applications of contrast ultrasound, as well as an evaluation of translational research regarding therapeutic applications of contrast ultrasound.

For this purpose, the thesis was divided in three parts. The first part focussed on the diagnostic application of contrast ultrasound in the field of cardiology and anaesthesiology. The second and third part of this thesis encompassed the translational research performed in the cardiovascular field of therapeutic application of contrast ultrasound. With the focus of the second part on in vivo studies performed and the third part evaluated its possible clinical application.

In part 1, chapter 2 reviewed the diagnostic application of contrast ultrasound for the quantitative assessment of myocardial perfusion in the perioperative setting. It concludes that microbubbles significantly enhance the blood compartment in ultrasound images thereby enhancing contrast ultrasound to a rapid and safe bedside technique with added clinical value. It also enables the qualitative and quantitative assessment of myocardial perfusion. This method has been shown to be accurate and may prove to be a powerful, non-invasive tool for studying the effects of anaesthesia, surgical procedures, and other perioperative factors on myocardial perfusion. In Chapter 3 the effect of general anaesthesia on myocardial perfusion in healthy subjects is assessed using diagnostic contrast ultrasound to quantitatively measure myocardial perfusion. Results showed that despite a decrease in myocardial blood volume at rest, myocardial perfusion was unaffected during sevoflurane anaesthesia. Furthermore, hyperaemic myocardial blood flow was reduced during anaesthesia, presumably caused by a decrease in perfusion pressure. Finally, sympathetic stimulation during sevoflurane resulted in a similar increase in myocardial blood flow compare with baseline. It remained to be elucidated whether these observed changes are due to the direct effect of sevoflurane on the myocardial vasculature, or a consequence of indirect hemodynamic alterations.

The second part discussed the results of therapeutic contrast ultrasound on arterial thrombosis in pig models. In chapter 4 two pig models were examined, a normal and an atherosclerotic model, in which ST elevation myocardial infarction was obtained by creating coronary thrombosis. The effect of high mechanical index contrast ultrasound on both microvascular flow and epicardial recanalization was evaluated. It was observed that guided high mechanical index impulses improved microvascular flow in peri-infarct segments, independent of epicardial recanalization. Larger reductions in perfusion defect size and epicardial recanalization rates were observed in the setting of hypercholesterolemia and underlying plaque. Furthermore, it
was concluded that the use of a three dimensional probe had a practical advantage in that high mechanical index impulses could be delivered to the entire risk area without probe manipulation. Chapter 5 delineated the set-up of a new pig model in which acute peripheral arterial occlusions was created and the treatment with contrast ultrasound was evaluated. We encountered severe anaphylactic reactions to the lipid shell of the microbubble used in our porcine model. This resulted in recommendations regarding the pre-treatment protocol using methylprednison intravenously, indomethacine suppository and aspirin intravenously. With the implementation of this protocol none of the remaining pigs experienced anaphylactic reactions. Results of contrast ultrasound on top of standard thrombolytic therapy in acute peripheral arterial occlusions in a pig model were discussed in chapter 6. It was observed that contrast ultrasound on top of thrombolysis, using urokinase, resulted in a significant reduction in thrombus weight. Furthermore, iliac blood flow, microcirculation, and limb arterial pressures tended to improve within 3 hours of therapy. On top of that, no haemorrhagic complications were encountered during these experiments. It was concluded that contrast ultrasound has the potential to improve thrombolytic therapy in large peripheral arterial occlusions and these results warrant further prospective studies in patients with peripheral arterial occlusions. The studies evaluating therapeutic application of contrast ultrasound in humans were discussed in part 3. It commenced with chapter 7 which outlined the rationale and the design of the Sonolysis study. This was worldwide the first study performed in ST elevation myocardial infarction patients evaluating the effect of contrast ultrasound on top of thrombolysis on epicardial recanalization. In chapter 8 results of the Sonolysis study were discussed. It demonstrated that the study protocol was safe and feasible and results warrants further inclusion of patients to increase the power of the study. Positive findings may stimulate future research and technical innovations to implement the treatment in the ambulance or in the emergency room when primary PCI is not readily available and potentially obtain patency at an earlier stage, resulting in smaller infarctions. Chapter 9 critically appraised the current literature with respect to therapeutic application of contrast ultrasound. It demonstrated that a considerable amount of research has been performed to demonstrate that contrast ultrasound enhances thrombolysis. Although the exact working mechanism remains to be elucidated, it is known that destruction of microbubbles and local application of ultrasound induces several bioeffects, resulting in enhanced thrombolysis. Animal studies also demonstrated that despite an absence of epicardial coronary recanalization, the peripheral perfusion of the area at risk did improve, indicating microvascular effects that are independent of upstream vascular flow and obstruction. This may be mediated by cavitation-induced activation of purinergic pathways, leading to prolonged increases in NO production that have a positive influence on the imbalance between NO and superoxide production and subsequent inflammatory responses. Pulse duration seemed to be playing a major role in
potential effects and side effects. It was concluded that further research is necessary in order to explore the potential application of contrast ultrasound in patients suffering microvascular obstruction after successful primary PCI. It was also discussed that high-intensity ultrasound proved its potential effect on thrombus dissolution. However, it has never been further investigated due to its invasive character. Recent technical developments made it possible to apply high-intensity ultrasound in a non-invasive manner. Pre-clinical research demonstrated that local application of high-intensity ultrasound in ischemic areas resulted in local coronary neovascularisation, thereby, potentially, reducing symptoms and ischemia in patients with refractory angina. In chapter 10 the results of the shockwave study are shown. This study demonstrated that local application of high-intensity ultrasound (shockwave therapy) improved NYHA functional class and reduced nitroglycerin use in our patient group. We also performed CMR to quantitively measure myocardial perfusion and subsequent ischemic burden. Nevertheless, we did not demonstrate any improvement in myocardial perfusion and ischaemic burden. Therefore, additional studies are necessary to understand the responsible working mechanism.

In summary, the work presented in this thesis described the diagnostic and therapeutic application of contrast ultrasound in cardiovascular disorders. It demonstrated that diagnostic contrast ultrasound is an excellent tool to measure myocardial perfusion in a bed-side manner. This creates the opportunity to perform future studies on the effect of anaesthesia, surgical procedures, and other perioperative factors on myocardial perfusion. Furthermore, pre-clinical and clinical studies demonstrated an effect on thrombus dissolution using contrast ultrasound on top of thrombolysis. This effect was observed on both macrovascular and microvascular level. These results warrant future studies to further evaluate the therapeutic potential of contrast ultrasound and its optimal settings. Additional technical developments also needs to be explored to make contrast ultrasound an easy-to-use therapy.