Ultrasound in Medicine & Biology 000 (2024) 1−11

Contents lists available at ScienceDirect

Ultrasound in Medicine & Biology

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

Original Contribution

Perioperative Detection of Cerebral Fat Emboli From Bone Using High-Frequency Doppler Ultrasound

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A R T I C L E I N F O

Keywords:
Bone marrow emboli
Fat embolism syndrome
Porcine model
Neonate
Cardiac surgery
Orthopedics
High-intensity transient signals (HITS)
Neurology
Doppler ultrasound

A B S T R A C T

Objective: Fat embolism syndrome and cerebral fat emboli are rare yet serious conditions arising from systemic distribution of bone marrow emboli. Emboli are known to produce high-intensity transient signals (HITS) in a Doppler signal. We hypothesized that both intramedullary nailing in pigs and median sternotomy in human infants cause bone marrow release, that some of these cause cerebral emboli, and that these were detectable by a new cerebral doppler ultrasound monitoring system (NeoDoppler). We also aimed to describe the intensity of HITS generated during these procedures.
Methods: Specific pathogen-free Norwegian landrace pigs were allocated to either bilateral femoral nailing or injection of autologous bone marrow (positive controls). Testing was carried out under continuous Doppler monitoring. Presence of cerebral emboli was confirmed with histology. NeoDoppler data from infants undergoing sternotomy prior to cardiac surgery were investigated for comparison.
Results: Eleven of twelve pigs were monitored with cerebral Doppler ultrasound during femoral surgery. HITS were seen in five (45%). Brain biopsies demonstrated bone marrow emboli in 11 of the 12 (92%). Four positive control pigs received intraarterial injections of bone marrow, saline, or contrast, and strong HITS were detected in all pigs (100%). Median sternotomy in eight human infants was associated with a significant increase in embolic burden; the HITS intensity was lower than HITS in pigs.
Conclusion: High-frequency cerebral Doppler ultrasound is a valuable tool for perioperative monitoring that can detect emboli in real-time, but sensitivity and specificity for bone marrow emboli may be limited and size-dependent.

Introduction

Fat embolism syndrome (FES) is a serious condition defined by the presence of fat globules in the systemic and pulmonary circulation [1]. The syndrome is commonly seen in trauma patients with long bone fractures or after orthopedic procedures but has also been reported following procedures such as liposuction and bone marrow biopsy [2]. Cardiac surgery, including sternotomy, in children is of special concern as cardiac shunts are common and may transport emboli directly into the systemic circulation [3]. In rare cases, cerebral symptoms present in isolation, a condition referred to as cerebral fat embolism (CFE). CFE can be challenging to diagnose and may result in the delay of appropriate treatment [4]; CFE may also occur in the absence of intracardiac shunt, such as *patent foramen ovale* [5]. Although magnetic resonance imaging (MRI) is considered the gold standard for CFE diagnosis [6,7], it is uncertain if early MRI may prove diagnostic.

Cerebral Doppler ultrasound can provide real-time detection of emboli in the cerebral circulation and may thus be used to quantify

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https://doi.org/10.1016/j.ultrasmedbio.2024.09.017
Received 23 June 2024; Revised 26 August 2024; Accepted 22 September 2024

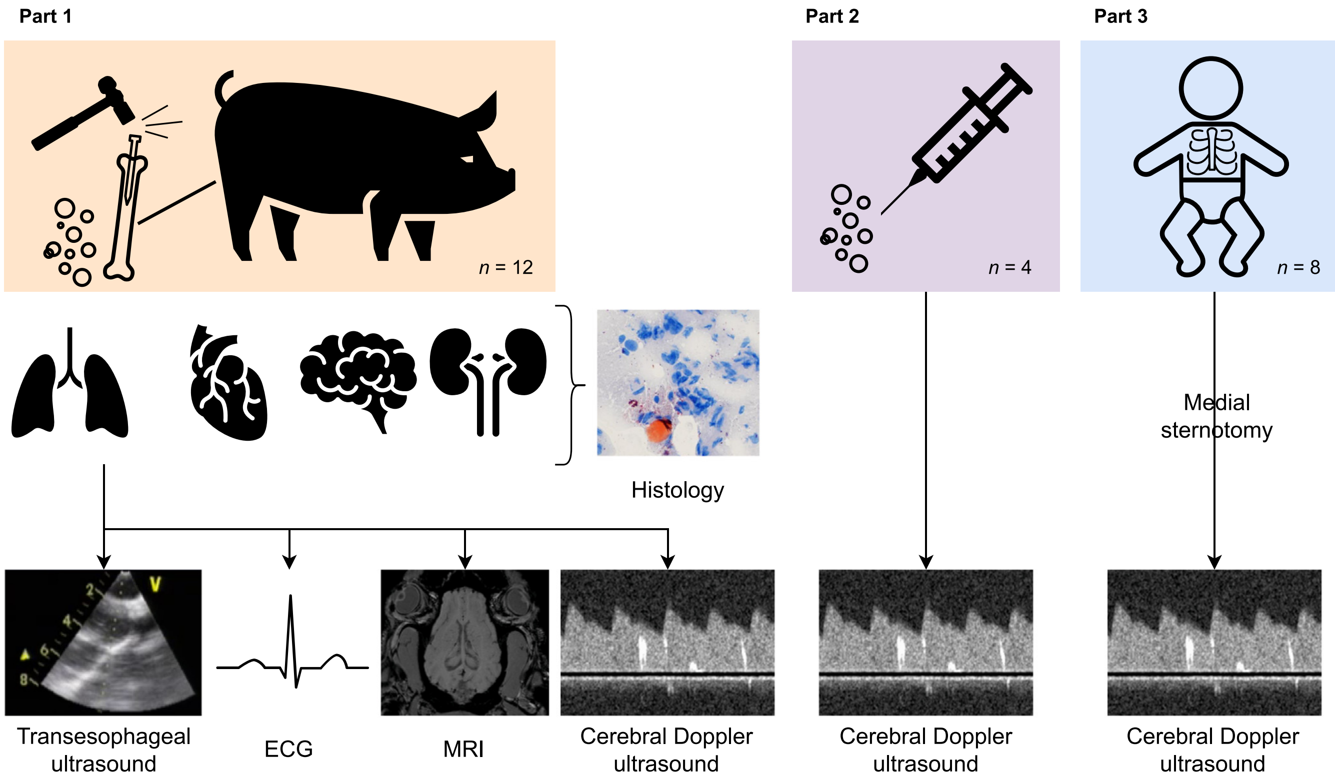

Figure 1. Overview of the methods used at each part of the study. Transesophageal echocardiography (TEE) was used to monitor lungs and heart, ECG was used to monitor the heart, cerebral doppler ultrasound and postmortem MRI were used to monitor the brain, and biopsies for histology were acquired from these locations as well as from kidney. Data from a separate study on infant open-heart surgery were included for comparison [3].

embolic load during medical procedures [8]. Solid or gaseous entities in the blood stream have higher reflectivity than surrounding erythrocytes and produce characteristic high-intensity transient signals (HITS) in the Doppler spectrum. Transcranial Doppler ultrasound has been used to detect HITS during various medical procedures [8], including orthopedic surgery [9] and cardiac surgery in infants [3]. The clinical significance of the presence of HITS has, however, not yet been established and there is a lack of studies correlating HITS characteristics with histologically verified brain emboli.

In this study, we aim to describe the relationship between surgically generated bone marrow emboli and ultrasound-detected HITS *in vivo*. To this end, we performed bilateral reamed intramedullary nailing of the femur in pigs without intracardiac shunt. Timing, presence, and extent of CFE was assessed by combining cerebral Doppler ultrasound monitoring, postmortem MRI, and histological examination of brain tissue. For validation, bone marrow was injected in the carotid artery of four additional animals with identical monitoring. Data from infant cardiac surgery were used for comparison.

Materials and methods

Part 1: HITS monitoring during femoral nailing in a porcine model

Study animals

Twelve specific pathogen-free Norwegian landrace pigs were assessed for inclusion in March−September 2022 [10]. Exclusion criteria were pre-existing disease, complications not caused by the femoral nailing itself, and *patent foramen ovale*. No animals were excluded prior to surgery.

Experimental procedure

A detailed protocol describing the surgery and anesthesia of this study has been published by Kristiansen et al. [10]. Data were collected through multimodal monitoring and postmortem examinations (Fig. 1). Biopsies were collected postmortem from lungs, heart, kidneys, and brain. The head was transferred to a nearby MRI facility and scanned within one hour of sacrifice.

Doppler ultrasound monitoring with the NeoDoppler system

The NeoDoppler ultrasound system was developed by the ultrasound group at NTNU and a research set-up was used in this study [11]. The single element plane wave transducer operates at a frequency of 7.8 MHz. It is a multigated Doppler, which makes it possible to measure blood flow velocities in several depths simultaneously down to approximately 35 mm. All depths are recorded, and the depth of interest is selected during analysis. Anatomical structures cannot be visualized as the set-up cannot produce B mode images and no angle correction is performed. Thermal and mechanical indices are kept within the recommended limits for long-term continuous monitoring, as verified with calibrated hydrophone measurements. The NeoDoppler ultrasound system can measure rapid changes in blood flow [12] and has been used to map the cerebral effects of general anesthesia [13] and detect HITS, trends and events [3,14] during cardiac surgery in infants. Data processing was performed with an in-house software developed in MATLAB (MathWorks© R2017a).

Trepanning was used to create a fontanel-like acoustic window to the cerebral blood flow. An L-incision was made, starting from the base of the ear to the midline, and perpendicular along the midline in anterior direction. The skull bone was then uncovered, and a circular opening with diameter of 2−3 cm was drilled with a Dremel 4000 multitool with flexible shaft and 6.4/7.9 mm Dremel carving bits (Dremel Europe, Breda, Netherlands) without penetrating the dura. The NeoDoppler probe was then placed on the dura and positioned so that a strong arterial signal was detected (Fig. 2). The probe was attached in place with a compress and a tourniquet. In cases where the carotid artery access was

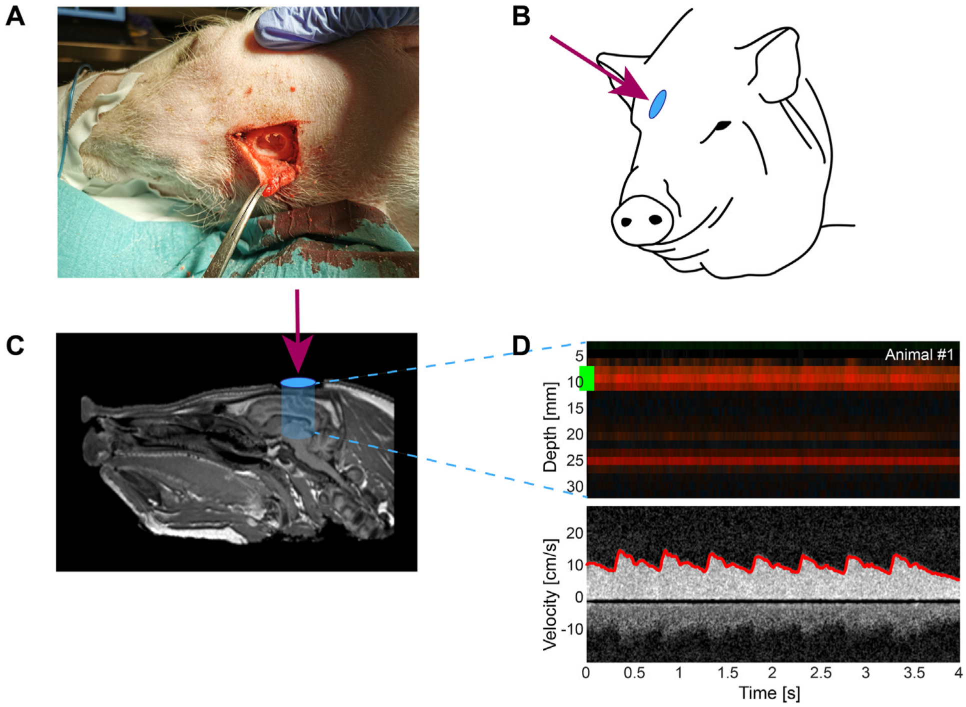

Figure 2. Positioning of the NeoDoppler probe. (**A, B**) A small opening was made in the skull without penetrating the dura, approximately in an L-incision starting at the base of the ear, continuing towards the midline, and then in anterior direction along the midline. The NeoDoppler probe was fixed in the opening as indicated by the purple arrow. (**C**) The opening in the skull was clearly visible on postmortem MRI images. The NeoDoppler probe insonates a cylindrical volume as indicated in blue. (**D**) The upper panel demonstrates color M mode showing the doppler signal at each depth of the blue cylinder. Blood approaching the probe is visualized in red and blood moving away from the probe is visualized in blue. Time on the *x* axis. The selected depth is shown as a green box. The lower panel shows the doppler signal of the corresponding volume, converted to a velocity spectrum. The velocity spectrum can be automatically traced, as shown in red.

established, the probe was positioned on the contralateral side. Otherwise, the right-hand side was chosen.

Histopathological analyses

Tissue samples of the brain were collected from four locations: dorsal and ventral at both brain hemispheres [10]. Serial sections from the brains of the two animals with most HITS during surgery were stained as follows: 12 µm unfixed frozen sections were stained in saturated Sudan III (Sudan Red III Merck 1380) in a mix of acetone and 70% ethanol (50:50) for 2 minutes and rinsed with running water. The sections were then counterstained with hematoxylin and coverslipped with an aqueous mounting media. Additional 8 and 12 µm unfixed cryosections were stained with the histological routine staining, hematoxylin-eosin-saffron, in an automatic slide stainer (Sakura Tissue-Tek © Prisma™). The slides were fixed and rehydrated through descending grades of ethanol to water before staining in hematoxylin (CellPath/Chemi-Teknik, RHD-1475-100, CI No 75290), followed by bluing in water and further staining with Erythrosine (239, VWR, no 720-0179). After rinsing in water for removal of excess dye, the sections were dehydrated through ascending grades of ethanol and stained in Saffron (Chemi-Teknic AS, Chroma 5A-394, CI No 75100), rinsed in several baths of absolute ethanol, and cleared in Tissue Clear before coverslipping in Sakura Tissue-Tek© Glas™ automatic coverslipper. The sections were dried overnight in a well-ventilated place to avoid chemical evaporation.

Sections were scanned on an Olympus VS200 at 20x magnification. The scanned images were analyzed in QuPath (v0.5.1), a software for digital histology [15]. Potential emboli were identified using a thresholder on the DAB (3, 3′-diaminobenzidine) channel with threshold 0.5, full resolution, and a lower limit of 1 µm^2 (Appendix A). The identified objects were annotated and reviewed manually. Artefacts and emboli outside the tissue section were excluded.

Data analysis

Doppler recordings were analyzed using in-house software to visualize the Doppler spectra and annotate HITS [3]. HITS occurring before start of surgery were excluded from further analysis. HITS intensity (the embolus-to-blood ratio, EBR) was calculated based on the difference in intensity between the manually marked single HITS and the mean background signal during the 5 s before and after all single HITS; HITS with curtain effect (large clusters of HITS where individual HITS cannot be distinguished) were excluded from analysis [3]. Statistical analyses were performed in R version 4.3.1 [16] and visualizations made with the ggplot2 and ggrain packages [17,18].

Part 2: HITS monitoring during carotid intraarterial bone marrow injections in a porcine model (positive control)

Study animals

Four additional specific pathogen-free Norwegian landrace pigs were assessed for inclusion in October 2022. Anesthesia and monitoring were established, as described above. No animals were excluded.

Experimental procedure

Bone marrow was aspirated from the tibia using the Arrow EZ-IO intraosseous cannula (Teleflex, Wayne, PA), mixed with heparin, and slowly injected venously. Towards the end of the monitoring period, bone marrow was again extracted from the tibia, mixed with heparin, and slowly injected into the carotid artery access line. This was repeated 4−6 times, interspersed with injections of agitated saline solution. The saline injections were intended to flush clean the arterial line and to act as a crude contrast agent to verify that the injected content reached the insonated area. Ultrasound contrast bubbles have, in contrast to agitated saline solution, a known bubble size distribution and were used as a reference. A clinically used ultrasound contrast agent (SonoVue microbubbles, Bracco Imaging, Italy) was prepared as per the manufacturer's instruction and

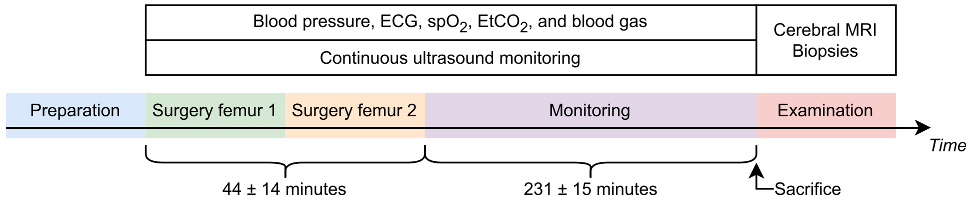

Figure 3. Stages of femoral surgery with mean ± SD duration in minutes. ECG, electrocardiogram; $EtCO_2$, end-tidal carbon dioxide level; spO_2, peripheral oxygen saturation.

administered to two animals. In the first animal, the contrast was injected after the first sequence of bone marrow and saline injections. The agent, however, proved stable and caused delay for further injections as it produced a strong signal for 20−30 minutes. Therefore, in the second animal, the contrast was administered at the end of the experiment. Data from two animals was considered enough as the agent produced characteristic patterns and seemed hemodynamically stressful to the animals by causing (temporary) pulmonal hypertension.

Part 3: HITS monitoring during sternotomy in human infants

Patient selection

Data have previously been collected from 28 infants below one year of age undergoing cardiac intervention at Oslo University Hospital, Norway recruited from February 2019 to December 2019, of which 13 underwent cardiac surgery with cardiopulmonary bypass [3]. The infants received NeoDoppler monitoring during various stages of the intervention. For the current study, data from infants being monitored during median sternotomy were included for reanalysis.

Experimental procedure and data analysis

The NeoDoppler probe was attached over the infant's fontanel and continuous recordings were made with 1 minute pauses [3]. HITS were manually annotated as described above. To examine if HITS intensity increased at the time of median sternotomy, the HITS detected in the period from five minutes before (t = -5) to five minutes after (t = 5) sternotomy (t = 0) were compared to similar ten-minute windows preceding (from t = -15 to t = -5) and

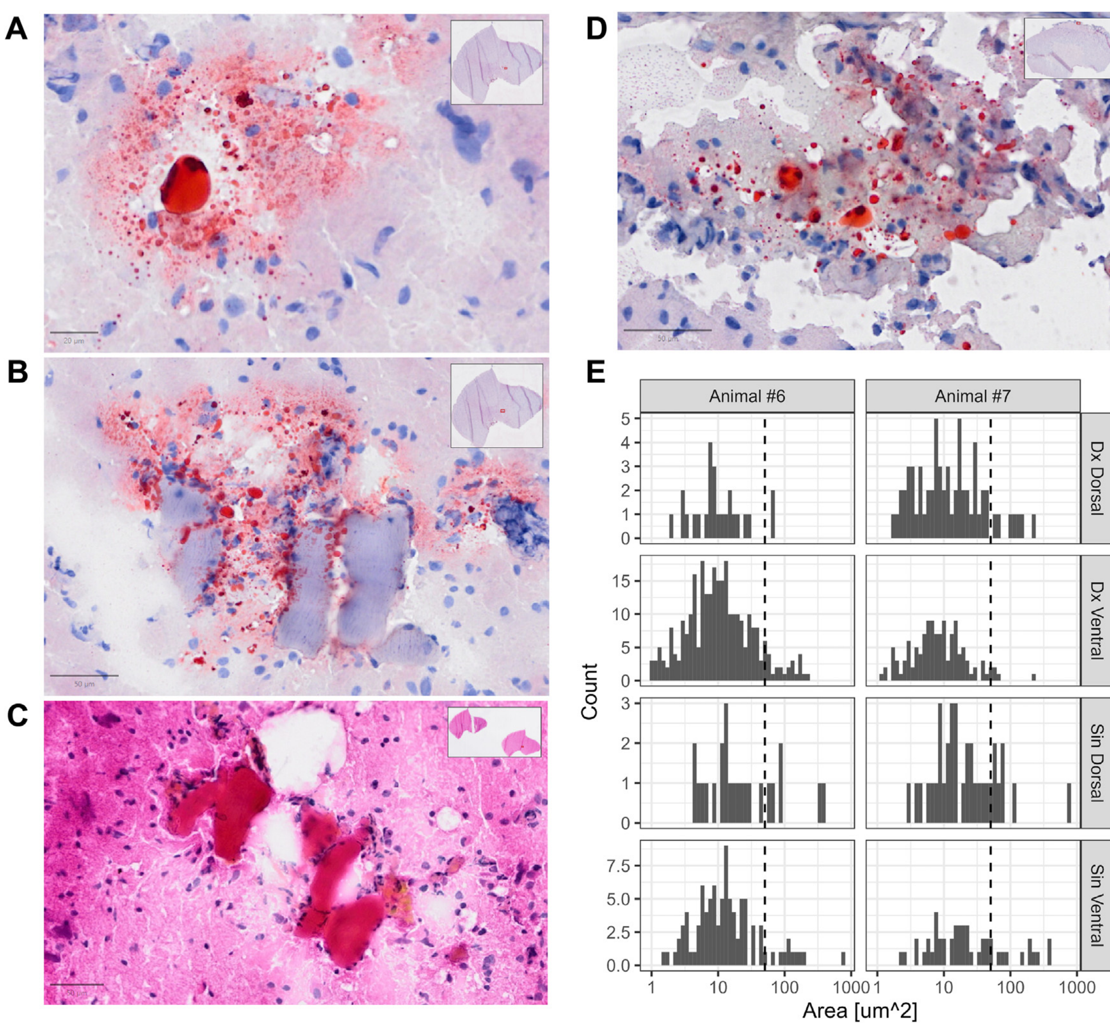

Figure 4. Histological verification of bone marrow emboli. (**A**) Example of a well-defined bone marrow emboli (26 × 19 μm) in animal #6 surrounded by diffused fat droplets, located in the brain parenchyma. (**B, C**) Bone fragments located in the brain parenchyma of animal #6, stained with (b) Sudan III and (c) H&E. (**D**) Bone marrow emboli located in meningeal tissue in animal #7. (**E**) Size distribution of emboli in the eight sections. The dashed line corresponds to the area of an erythrocyte (approximately 50 μm^2). Abbreviations: Dx, dexter (right-hand side); Sin, sinister (left-hand side).

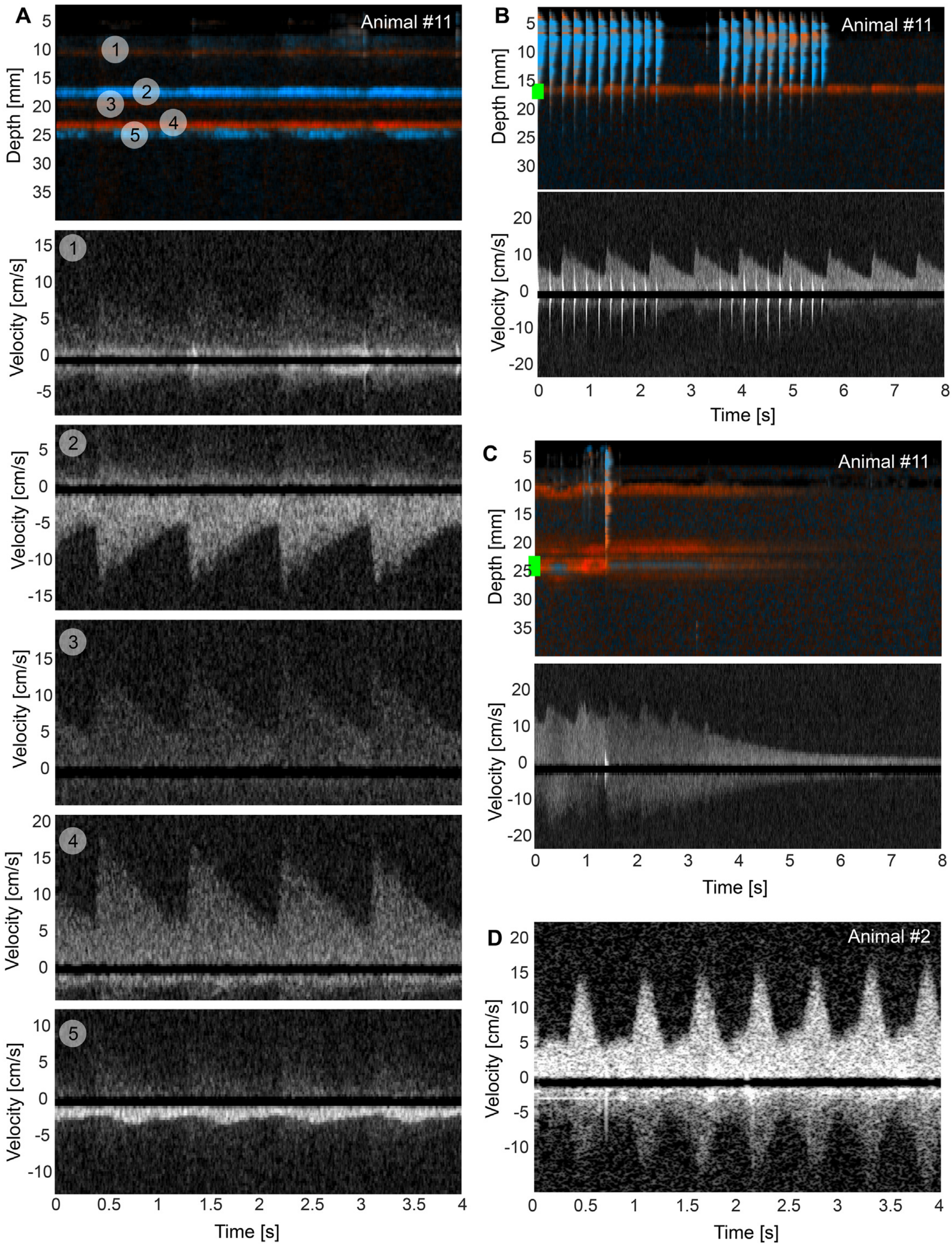

Figure 5. Doppler spectra from animals included in part 1 of the study. Upper panels (color M mode) show the depth of the selected signal (numbered or green box). (**A**) Signal from multiple depths, indicated as 1−5 in the upper panel (M mode). The fifth depth shows a characteristic venous signal. (**B**) Impact of hammering on the Doppler spectrum. (**C**) Cessation of pulsatile cerebral blood flow at multiple depth at sacrifice. (**D**) Inverted diastolic blood flow during sacrifice.

following (from $t = 5$ to $t = 15$). First, HITS intensity between periods were compared using the Krusk−Wallis test and Dunn's test with Benjamini−Hochberg correction for pairwise multiple comparisons. Second, linear mixed models were used to take interindividual differences into account by including participant as random intercept. p values were estimated with Satterthwaite's method using the lmerTest package for R [19].

Ethics

Parts 1 and 2 were approved by the Norwegian Animal Research Authority (FOTS ID 19803), and the studies adhered to the Norwegian Laboratory Animal Regulations and the EU directive 2010/63/EU. Part 3 was approved by the Regional Committee for Medical and Health Research Ethics, REC Central (Reference 2017/314), the Norwegian

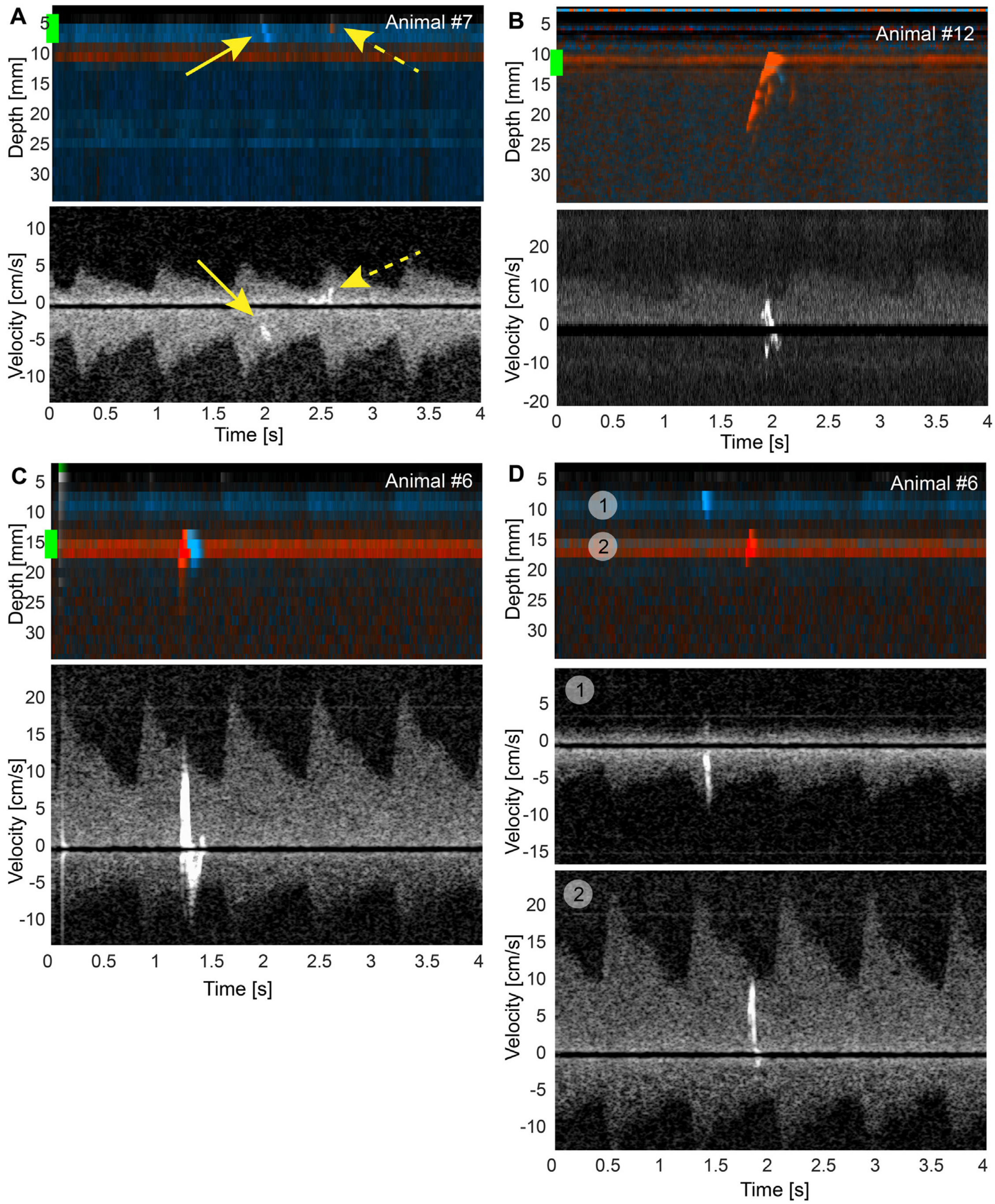

Figure 6. Doppler spectra from animals included in part 1 of the study. Strong high-intensity transient signals (HITS) following femoral surgery in three different animals (**A−D**).

Directorate of Health (Reference 17/15181-11) and The Norwegian Medicines Agency (Reference 19/05458), and informed written consent was obtained from the parents of all participants.

Results

Part 1: HITS monitoring during femoral nailing in a porcine model

Experimental characteristics

Bilateral reamed intramedullary nailing of the femur was performed on 12 pigs (median weight 29.5 kg, 11 males, Table S1) as reported by Kristiansen et al. [10]. Mean ± SD experimental duration from start of surgery to sacrifice was 275 ± 18 minutes (Fig. 3).

Cerebral fat embolism from reamed intramedullary nailing of the femur

Cerebral bone marrow emboli were identified in brain biopsies from 11 of 12 animals and systemic fat emboli were demonstrated in biopsies from lungs and heart in all animals as previously reported [10]. Brain tissues from the two animals with most HITS were restained. In total, 700 emboli were found across eight brain tissue sections (Fig. 4), with median and maximum areas 11 μm^2 and 795 μm,^{2} respectively. Only 11% of the emboli were larger than an erythrocyte (Fig. 4D). The emboli were primarily located in or close to the meninges but could also be found in the parenchyma. In addition, bone fragments were found in two sections (Fig. 4C).

Real-time detection of cerebral emboli using NeoDoppler

Monitoring of cerebral blood flow using the Doppler ultrasound based NeoDoppler technology was successfully established in 11 of 12 experiments. One experiment was conducted without NeoDoppler monitoring due to perforation of the *dura mater* during the drill during preparation. Recordings were graded as acceptable, partly acceptable, and unacceptable, based on visual evaluation of signal quality and signal stability. In total, 2847 minutes of Doppler ultrasound recordings were collected, of which 2181 (77%), 651 (23%), and 14 (<1%) minutes were acceptable, partly acceptable, and unacceptable recordings, respectively (Fig. S1). Partly acceptable recordings contained shorter periods with very weak or absent doppler signals, most often due to positional manipulation of the pig.

High-quality Doppler signals were often available for more than one depth (Fig. 5A) and instantly reflected the interventions in various stages of the experiments, such as diathermy usage, reaming, nailing (Fig. 5B), and sacrifice (Fig. 5C,D). Furthermore, episodes with arrhythmia or acute hypotension were also clearly visible in the Doppler signal.

Perioperative HITS were observed in seven of the eleven monitored animals (Figs. 6 and S1). Notably, the first animal exhibited HITS before start of surgery, reflecting a routine arterial line flush to prevent clotting. These HITS were thus excluded from further analysis.

Part 2: HITS monitoring during carotid intraarterial bone marrow injections in a porcine model (positive control)

Experimental characteristics

As positive controls, four additional pigs (median weight 29 kg, 4 males, Table S1) received venous injections of autologous bone marrow instead of surgery. Mean ± SD time from first injection to sacrifice was 262 ± 8 minutes. Before sacrifice, additional intraarterial injections of bone marrow, agitated saline solution and ultrasound contrast were administered during NeoDoppler monitoring.

The NeoDoppler ultrasound system successfully detects bone marrow emboli

Strong HITS were observed in all intraarterial injection experiments. Injections were made contralaterally to the NeoDoppler probe in two of the injection experiments due to unexpected difficulties gaining carotid access. The first animal received undiluted bone marrow in the carotid artery contralaterally to the NeoDoppler probe. Strong HITS were observed immediately after injection (Fig. 7A). During injection, the Doppler signal suddenly disappeared (Fig. 7B) and we were unable to find a new signal. As the probe was not moved during injection, this suggests actual cessation of blood flow, possibly caused by occlusion of proximal arteries. Subsequent animals received diluted bone marrow (1:2 and 1:4) and did not experience occlusion (Fig. 7C−E).

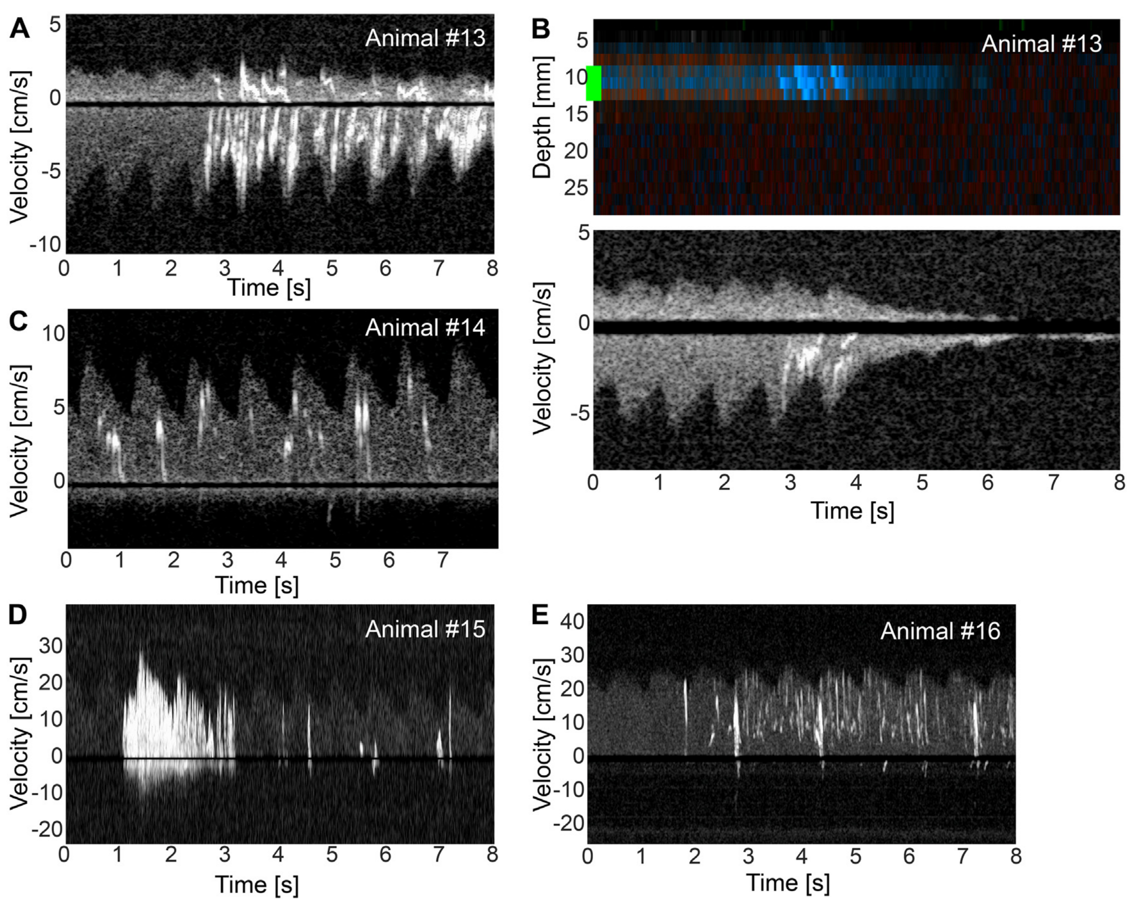

Figure 7. Strong high-intensity transient signals (HITS) following injection of autologous bone marrow into the carotid artery of four different animals (A, **C**−**E**). (**B**) Sudden cessation of blood flow following injection of autologous bone marrow into the contralateral carotid artery. The upper panel (color M mode) shows the depth of the selected signal (green box). The HITS are visible as angled lines in M mode.

HITS characteristics of solid and gaseous emboli

Cerebral Doppler ultrasound recordings were made during controlled arterial injection of bone marrow and agitated saline solution in four pigs. In addition, ultrasound contrast was given to two of these animals.

Visually, the three injected agents produced characteristic HITS in the Doppler spectrum (Fig. 8). Injections of saline solution produced continuous short-lived curtains that sometimes separated into individual HITS. The initial signal from bone marrow injection was similar to injection of saline solution, but almost always transitioned from curtains into more or less separate HITS. Ultrasound contrast produced a distinct pattern with fine-grained HITS and lower intensity. Injections of saline solution seemed to produce HITS with higher intensity than injections of autologous bone marrow (Fig. 9), but the difference was not significant when adjusting for animal with linear mixed methods ($p = 0.645$).

Part 3: HITS monitoring during sternotomy in human infants

Study characteristics

Thirteen infants undergoing cardiac surgery with simultaneous NeoDoppler monitoring had annotated timing of median sternotomy [3]. Median weight and age at the time of the procedure were 6.2 kg (IQR: 4.0−6.5 kg) and 3.5 months (IQR: 2.8−5.0 months), respectively, and eight of the thirteen (62%) were male. HITS were observed during sternotomy in eight infants (62%, Table S1), who were included for further analysis.

HITS load and intensity

Two infants showed a particularly high embolic load during sternotomy (Fig. 10). The infant with the highest HITS count ($n = 44$) had transposition of the great arteries with intact ventricular septum, previously treated with balloon septostomy, operated with arterial switch. The infant with the second highest HITS count ($n = 24$) was a surgical repair of a transitional atrioventricular septal defect.

Sternotomy was associated with a significant increase in HITS burden. The number and intensity of HITS were higher around sternotomy than before and after ($H(2) = 37.14$, $p < 0.001$, Fig. 10). Higher HITS intensity during sternotomy compared to pre ($\beta = -3.260$, $p < 0.05$) but not post ($\beta = -2.257$, $p = 0.087$) was demonstrated using linear mixed models to take interindividual differences into account.

The intensity of surgery-generated HITS from the porcine model and neonates are shown in Figure 9. HITS from neonatal sternotomy had lower intensity than those obtained from the porcine model ($p < 0.001$).

Discussion

In this study, we demonstrate that the NeoDoppler ultrasound system can detect bone marrow emboli *in vivo* both during intra-arterial injection and during reamed intramedullary nailing of the femur in a porcine model. HITS produced during femoral surgery in pigs have different

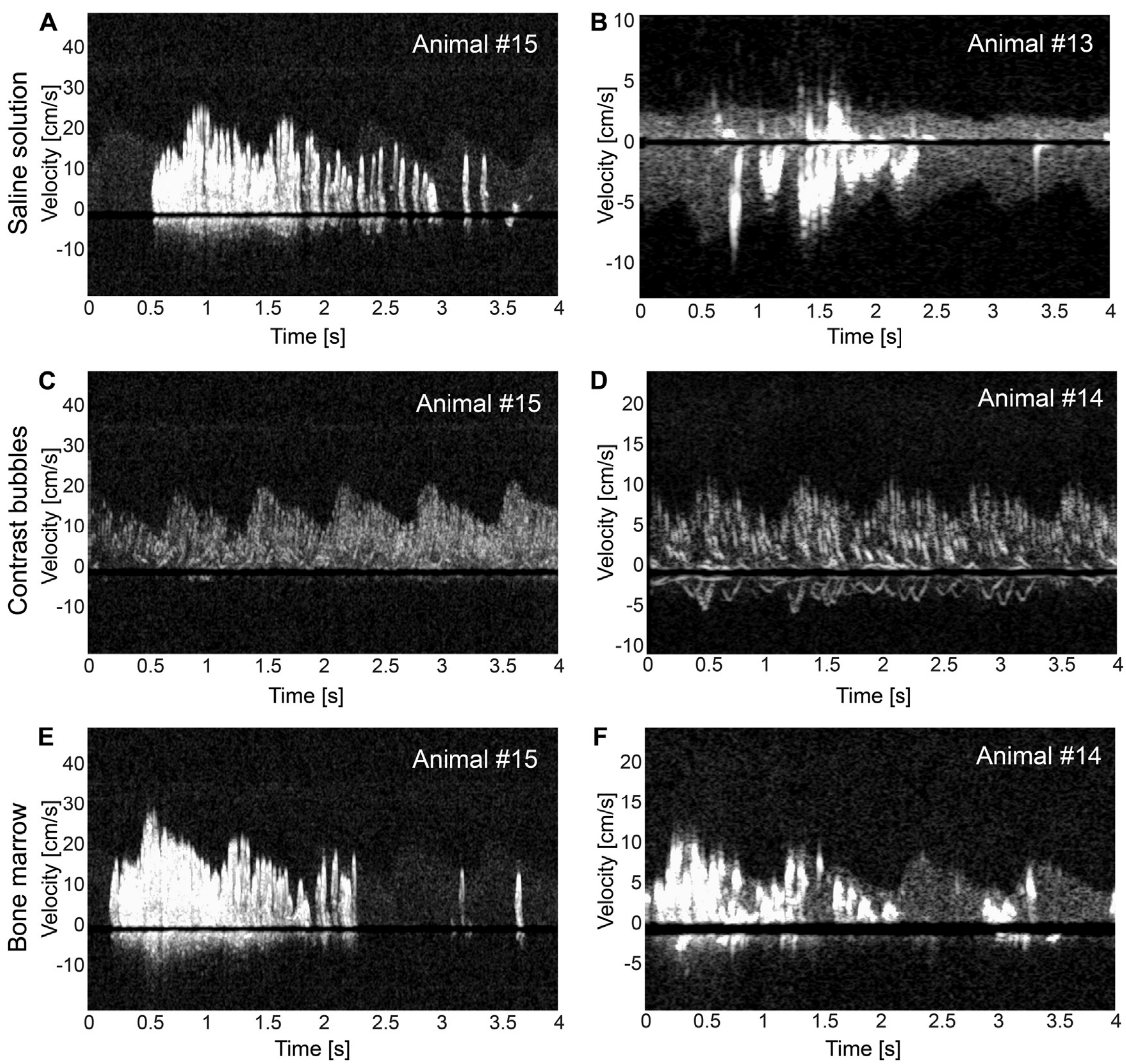

Figure 8. Examples of HITS produced by injection of (**A, B**) agitated saline solution, (**C, D**) ultrasound contrast, and (**E, F**) bone marrow.

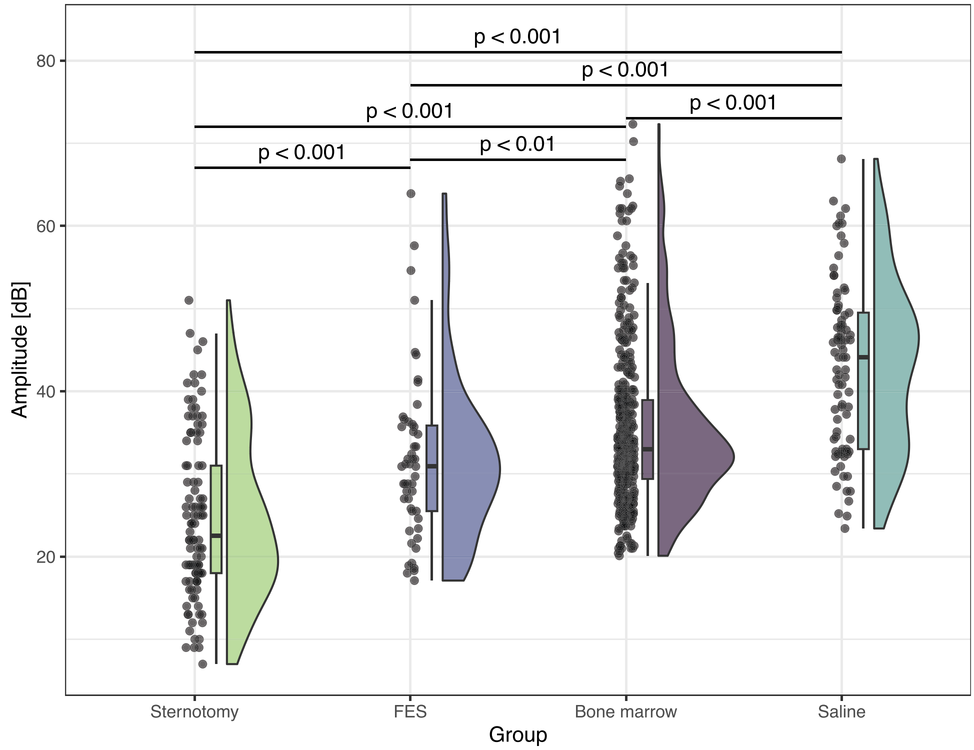

Figure 9. Comparison of high-intensity transient signals (HITS) intensity across exposure. HITS from sternotomy ($n = 104$) in human neonates had lower intensity than HITS from pigs undergoing femoral surgery (FES, $n = 51$) and control animals receiving injections of saline solution (saline, $n = 85$) and autologous bone marrow ($n = 394$).

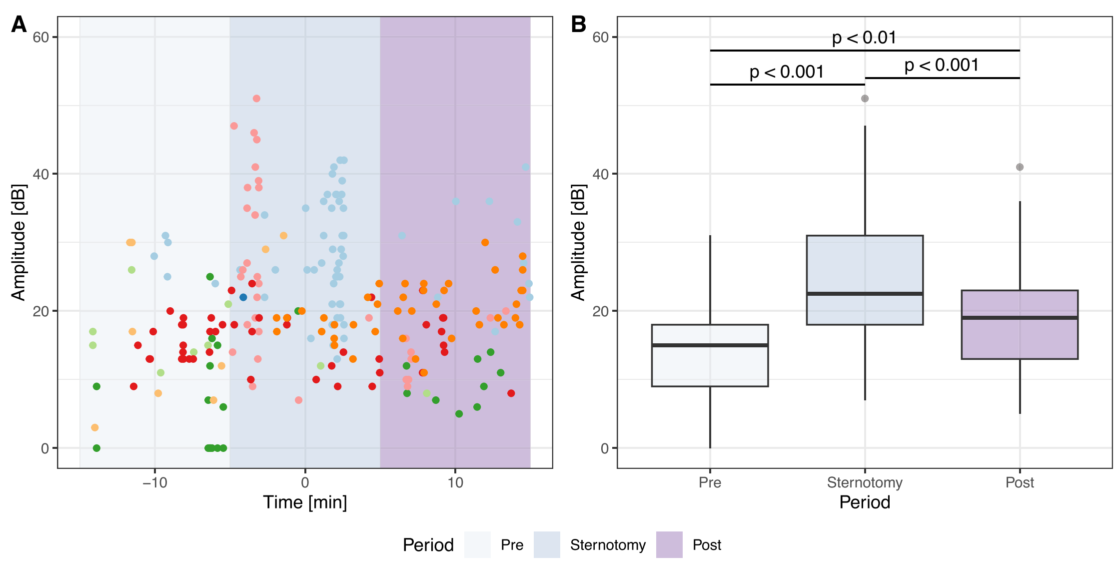

Figure 10. High-intensity transient signals (HITS) recorded in human infants during cardiac surgery. (**A**) HITS occurring in three subsequent periods of 10-minutes were compared. In total, 53, 104, and 63 HITS were detected in the pre, sternotomy, and post periods, respectively. The HITS are colored by individual. (**B**) HITS intensity was significantly different between periods. Adjusted *p* values from Dunn's rank sum test.

intensity compared to HITS produced during median sternotomy in human infants. Although TEE was able to visualize emboli real-time in the pulmonal artery in the porcine model, systemic emboli were not detected with this method [10]. Cerebral Doppler ultrasound successfully detected systemic emboli but with limited sensitivity in the described porcine model, possibly related to the small emboli size.

Although Doppler ultrasound has been available for perioperative emboli detection for many years [9,20], our literature search has not found previous studies validating the connection between cerebral HITS and histologically verified bone marrow emboli. The NeoDoppler probe operates at high frequency which is less sensitive to larger emboli than lower frequencies [21]. EBR decreases with increasing probe frequency, especially for larger emboli [22]. Thus, most studies on emboli detection apply a probe operating at about 2 MHz which is also well-suited to penetrate the bone of the skull in adults. The true embolic load is likely to be higher than what is estimated with the NeoDoppler system as smaller emboli are not detectable, but the clinical impact of these undetected emboli is difficult to evaluate. For example, Forteza et al. [23] studied patients with femoral shaft fracture and found neurological dysfunction only in patients with concurrent right-to-left shunt despite cerebral HITS being detected in all included patients. Manual annotation of HITS may lead to underestimation of emboli with low EBR, but the intra-arterial injection of bone marrow demonstrated strongly visible signals. Our finding that single-frequency Doppler ultrasound is unable to separate solid and gaseous emboli agrees with existing literature [24]. However, some differences in intensity were seen. Both kinds of emboli are likely to develop during femoral surgery [25]. The development of proper algorithms for HITS detection and classification is ongoing [26].

The intensity and number of HITS during pediatric cardiac surgery were different from the HITS generated by bone marrow in pigs. The lower number of HITS observed during femoral surgery may be related to anatomical filters such as the lungs, which are bypassed in infants with shunts or transposition of the great arteries. Furthermore, the *retia mirabilia,* the communicating vascular meshes located between the internal carotid arteries and ascending pharyngeal arteries in pigs [27], is not found in humans. Hence, the HITS frequency in the porcine model is not completely similar to human surgery [28]. In contrast, the infant with highest HITS counts during sternotomy had transposition of the great arteries, where venous blood is distributed systemically without passing through the lungs. Most emboli identified by histological staining were, however, smaller than ordinary erythrocytes and are likely to have passed through the lungs, but the clinical impact of such small emboli is uncertain. The stronger intensity of HITS observed in pigs compared to the infants may reflect differences in bone marrow composition. Neonatal bone marrow is almost free of lipids and consists of hematopoietic elements [29], but the red bone marrow is replaced by fatty marrow with advancing age. At 20 years of age, most of the long bone marrow has been converted to fatty marrow [30]. The increasing lipid content in bone marrow with advancing age has also been seen in pigs [31]. Importantly, the pathophysiology of FES includes more elements than just emboli, such as immune activation and endothelial dysregulation [32]. Acute kidney injury may be another mediator that causes hemodynamic instability and brain injury [33−35]. It is also tempting to speculate whether this difference in bone marrow composition may impact the immunological response to the emboli. FES and CFE are traditionally believed to be rare in children [36], but CFE has been found in 15% of pediatric autopsies, even in nontraumatic deaths and despite absence of *patent foramen ovale* and pulmonal fat emboli [37]. Thus, intracardiac shunt does not appear to be a necessary prerequisite to develop systemic embolization and CFE, consistent with our findings in the porcine experiments.

Data on the embolic load associated with specific medical procedures can be clinically valuable before, during and after surgery. Preoperatively, such data may help the surgical team evaluate the risk of a procedure for a specific patient and to select the safest approach. For example, patients with *patent foramen ovale* may require special attention when procedures with high embolic load are considered [9]. During surgery, real-time feedback on embolic load may allow the surgical team to dynamically adjust the procedure to reduce the embolic load. Postoperatively, data on the actual embolic load experienced could inform the prognosis and guide diagnostic evaluation if complications arise [38]. There is, however, a need for further studies and standardization to fully understand the clinical usefulness of Doppler monitoring. Validated, reproducible techniques for quantifying embolic load are required to enable meaningful comparisons across procedures and patients.

Our study has several limitations. The study size was based on power calculations for the primary goal of confirming that femoral reaming and nailing can cause systemic emboli [10]. Male sex was clearly overrepresented in this study. However, the increased risk of FES among males is believed to be caused by the risk and mechanism of trauma rather than biological differences [36]. A sternotomy procedure is rather short, but a ten minutes window was used as the timing of procedure to make sure the annotated period was included for all infants and it is uncertain for how long the potential embolization continues after the end of the procedure. As can be seen in Figure 9, there is a tendency to higher HITS intensity in the first epoch after sternotomy than the epoch before sternotomy. However, other surgery related procedures could also influence this result. Finally, one of the limitations of single element Doppler ultrasound is the inability to identify anatomical structures and select specific blood vessels for examination. The detection of HITS during injection in the carotid artery contralateral to the NeoDoppler may thus be explained by both cerebral collaterals and insonation of several or contralateral cerebral vessels. Longitudinal studies are needed to evaluate the clinical impact of surgery-generated HITS.

Conclusions

Femoral reaming and nailing can produce bone marrow emboli that may reach the brain, even in absence of intracardiac shunt in pigs. High-frequency transfontanellar ultrasound is able to detect HITS generated by such emboli but with limited sensitivity in pigs. Sternotomy in human infants increased the number and intensity of HITS which may reflect increased embolic burden during bone manipulation.

Conflict of interest

NTNU and St. Olavs Hospital, Trondheim University Hospital may benefit financially from a commercialization of NeoDoppler used for cerebral Doppler monitoring in infants through future possible intellectual properties. H.T. and S.A.N are coinventors of NeoDoppler. S.A.N. is a board member of CIMON Medical. H.T. and S.A.N. have part-time positions and are among shareholders in CIMON Medical, the company responsible for commercialization of NeoDoppler. A.H.J., S.K., M.L.−O., C.V., I.N., and E.W.N. declare no conflicts of interest.

Data Availability statement

Raw data are available from the corresponding author, A.H.J., upon reasonable request. Please note that privacy regulations apply to data involving human participants.

Acknowledgments

Part 1 and 2 of the study received funding from Helse Nord, the Odd Berg Group, The Royal Norwegian Society of Sciences and Letters, and the Blix Family Fund. Part 3 received financial support from the Research Council of Norway (Commercializing of R&D [FORNY] program), the The Liaison Comittee for Education, Research and Innovation in Central Norway (References 2017/38297, 2018/42794, 2019/38881 and 2023-32976), The Norwegian Association for Children with Congenital Heart Disease's Research Foundation and The Heart Foundation, St. Olav's University Hospital's gift foundation. The histological staining was performed at the Histology lab at the Cellular and Molecular Imaging Core Facility (CMIC), Norwegian University of Science and Technology (NTNU). CMIC is funded by the Faculty of Medicine at NTNU and Central Norway Regional Health Authority. We would like to thank associated professor emerita Nancy Eik-Nes for proofreading the English language of this article.

Supplementary materials

Supplementary material associated with this article can be found in the online version at doi:10.1016/j.ultrasmedbio.2024.09.017.

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