EFSUMB Technical Review – Update 2023: Dynamic Contrast-Enhanced Ultrasound (DCE-CEUS) for the Quantification of Tumor Perfusion

Technisches Review der EFSUMB – Update 2023: Dynamischer kontrastverstärkter Ultraschall (DCE-US) zur Quantifizierung der Tumorperfusion

Authors

Christoph F. Dietrich[1, 2], Jean-Michel Correas[3, 4, 5], Xin-Wu Cui[6], Yi Dong[7], Roald Flesland Havre[8, 9], Christian Jenssen[10, 11], Ernst Michael Jung[12], Martin Krix[13], Adrian Lim[14], Nathalie Lassau[15], Fabio Piscaglia[16, 17]

Affiliations

1 Department General Internal Medicine, Kliniken Hirslanden Beau Site, Salem und Permanence, Bern, Switzerland
2 Zentrum der Inneren Medizin, Johann Wolfgang Goethe Universitätsklinik Frankfurt, Frankfurt, Germany
3 Department of Adult Radiology, Assistance Publique Hôpitaux de Paris, Necker University Hospital, Paris, France
4 Paris Cité University, Paris, France
5 CNRS, INSERM Laboratoire d'Imagerie Biomédicale, Sorbonne Université, Paris, France
6 Department of Medical Ultrasound, Tongji Hospital, Tongji Medical College, Huazhong University of Science and Technology, Wuhan, China
7 Department of Ultrasound, Xinhua Hospital Affiliated to Shanghai Jiaotong University School of Medicine, Shanghai, China
8 Department of Medicine, National Centre for Ultrasound in Gastroenterology, Haukeland University Hospital, Bergen, Norway
9 Department of Clinical Medicine, University of Bergen, Bergen, Norway
10 Department of Internal Medicine, Krankenhaus Märkisch Oderland Strausberg/ Wriezen, Wriezen, Germany
11 Brandenburg Institute for Clinical Ultrasound (BICUS), Medical University Brandenburg, Neuruppin, Brandenburg, Germany
12 Institute of Diagnostic Radiology, Interdisciplinary Ultrasound Department, University Hospital Regensburg, Regensburg, Germany
13 Global Medical & Regulatory Affairs, Bracco Imaging, Konstanz, Germany
14 Department of Imaging, Imperial College London and Healthcare NHS Trust, Charing Cross Hospital Campus, London, United Kingdom of Great Britain and Northern Ireland
15 Imaging Department. Gustave Roussy cancer Campus. Villejuif, France. BIOMAPS. UMR 1281. CEA. CNRS. INSERM, Université Paris-Saclay, France
16 Division of Internal Medicine, Hepatobiliary and Immunoallergic Diseases, IRCCS Azienda Ospedaliero-Universitaria di Bologna, Bologna, Italy
17 Dept of Medical and Surgical Sciences, University of Bologna, Bologna, Italy

Key words

contrast-enhanced ultrasound, guidelines, quantification, therapy monitoring, tumor perfusion

received 04.04.2023
accepted after revision 15.08.2023
published online 25.09.2023

Bibliography

Ultraschall in Med 2024; 45: 36–46
DOI 10.1055/a-2157-2587
ISSN 0172-4614

Georg Thieme Verlag KG, Rüdigerstraße 14, 70469 Stuttgart, Germany

Correspondence

Prof. Christoph F. Dietrich, MD
Department General Internal Medicine, Kliniken Hirslanden Beau Site, Salem und Permanence, Schänzlihalde 11, 3036 Bern, Switzerland
ChristophFrank.Dietrich@hirslanden.ch

ABSTRACT

Dynamic contrast-enhanced ultrasound (DCE-US) is a technique to quantify tissue perfusion based on phase-specific enhancement after the injection of microbubble contrast agents for diagnostic ultrasound. The guidelines of the European Federation of Societies for Ultrasound in Medicine and Biology (EFSUMB) published in 2004 and updated in 2008, 2011, and 2020 focused on the use of contrast-enhanced ultrasound (CEUS), including essential technical requirements, training, investigational procedures and steps, guidance regarding image interpretation, established and recommended clinical indications, and safety considerations. However, the quantifica-

tion of phase-specific enhancement patterns acquired with ultrasound contrast agents (UCAs) is not discussed here. The purpose of this EFSUMB Technical Review is to further establish a basis for the standardization of DCE-US focusing on treatment monitoring in oncology. It provides some recommendations and descriptions as to how to quantify dynamic ultrasound contrast enhancement, and technical explanations for the analysis of time-intensity curves (TICs). This update of the 2012 EFSUMB introduction to DCE-US includes clinical aspects for data collection, analysis, and interpretation that have emerged from recent studies. The current study not only aims to support future work in this research field but also to facilitate a transition to clinical routine use of DCE-US.

ZUSAMMENFASSUNG

Der DCE-US (Dynamic contrast-enhanced ultrasound) ist eine Quantifizierungstechnik des kontrastverstärkten Ultraschalls. Die EFSUMB-Leitlinien von 2004, mit Updates aus den Jahren 2008, 2011, 2013 und 2020, erläutern die Grundlagen der Ultraschall-Kontrastmitteltechniken, geben aber keine detaillierten Informationen zu den Anwendungsmöglichkeiten, der Vorgehensweise und den Besonderheiten des DCE-US. Ziel dieses EFSUMB-Dokuments ist es nun, auf der Basis einer aktuellen Literaturrecherche Standardisierungsgrundlagen zur Methodik des DCE-US – insbesondere für das Therapiemonitoring bei onkologischen Erkrankungen – weiter zu vertiefen. Die notwendigen Grundlagen und technischen Voraussetzungen für die Analyse von Zeit-Intensitätskurven werden vorgestellt. Das vorliegende Update eines EFSUMB-Statements aus dem Jahr 2012 berücksichtigt klinische Aspekte aufgrund jüngster Studien für einen standardisierten Ablauf der Daten-Akquise und -Analyse sowie Empfehlungen zur Interpretation. Die aktuelle Arbeit zielt nicht nur darauf ab, künftige Arbeiten auf diesem Forschungsgebiet zu unterstützen, sondern auch den Übergang zur klinischen Routineanwendung des DCE-US zu erleichtern.

Introduction

Dynamic contrast-enhanced ultrasound (DCE-US) is a technique to quantify tissue perfusion down to the capillary level based on phase-specific enhancement after injection of microbubble contrast agents for diagnostic ultrasound. In addition, the quantitative analysis of the dynamics of contrast enhancement overcomes its subjective comparison between normal and abnormal parenchyma, or between a focal lesion and the surrounding tissue.

The guidelines of the European Federation of Societies for Ultrasound in Medicine and Biology (EFSUMB) published in 2004 [1] and updated in 2008 [2], 2012 [3], 2013 [4, 5], and 2020 [6, 7] focused on the use of contrast-enhanced ultrasound (CEUS), including essential technical requirements, training, investigational procedures and steps, guidance regarding image interpretation, established and recommended clinical indications, and safety considerations. However, the quantification of phase-specific tissue enhancement acquired with ultrasound contrast agents (UCAs) is not discussed. The basis for the standardization of DCE-US has been established and published by the EFSUMB introductory paper in 2012 [3]. It provided some recommendations and descriptions of the quantification of DCE-US images, and technical explanations for the analysis of time-intensity curves (TICs).

As part of the development of professional standards for diagnostic ultrasound techniques [8] and in accordance with the regulations for EFSUMB policy documents published in 2019 [9], the current update was prepared on the basis of an up-to-date literature search. It includes clinical aspects for data collection, analysis, and interpretation in the quantification of tumor perfusion, which are derived from recent studies. This study focuses on the clinical assessment in oncology, but the basic considerations are generally transferable to other DCE-US indications such as treatment monitoring in inflammatory bowel disease or chronic kidney disease. The current study not only aims to support future work in this research field but also to facilitate a transition to clinical routine use of DCE-US.

Why do we need quantification?

Quantification of CEUS is needed to evaluate data objectively, to enable comparison of imaging techniques, to evaluate new UCA applications, to quantify tissue and tumor enhancement in order to characterize focal lesions, to evaluate therapeutic response, and to limit variability in clinical diagnosis [3]. Tissue perfusion is a relevant functional imaging parameter with pathophysiological and clinical relevance in different clinical settings and can be assessed with different imaging techniques, e. g., brain perfusion in stroke imaging using magnetic resonance imaging (diffusion) or dynamic contrast-enhanced computed tomography or myocardial perfusion using dynamic contrast-enhanced echocardiography for the heart.

An objective and quantitative diagnosis of perfusion characteristics is of particular relevance in the follow-up of cancer patients but can also be used for the diagnostic assessment of other pathological changes associated with alterations in tissue perfusion. This applies, for example, to the noninvasive diagnosis of the progression of parenchymal liver disease, liver cirrhosis, and portal hypertension [10, 11, 12, 13, 14, 15, 16] and for the noninvasive evaluation of chronic kidney disease [17, 18, 19] and subclinical kidney transplant rejection [17, 20, 21, 22, 23, 24]. There are partially contradictory data regarding the evaluation of inflammatory activity and response to biologic therapy in inflammatory bowel disease [25, 26, 27, 28, 29, 30, 31, 32, 33, 34]. A relatively new field of research is the application of DCE-US for the differential diagnosis, grading of the biological behavior, and outcome assessment of malignant tumors [35, 36, 37, 38, 39, 40, 41]. This position paper is focused on the assessment of tumor perfusion.

DCE-US as a dynamic examination is based on relatively long video sequences that measure changes in contrast signal over time from the bolus transit in the body. For precise diagnostic evaluation, such data need to be analyzed to extract biomarkers and other parameters that are related to relevant physiologic and

patho-physiologic properties and presented in a form that is compatible with the imaging process (e. g., color coded maps). It may be anticipated that such quantitative measures may play a major role in big data analysis and the development of machine learning, which itself may influence diagnostic approaches. Thus DCE-US has the potential to strengthen the role of CEUS in future diagnosis and follow-up [42, 43].

Current assessment of response to cancer treatment is still mainly based on interval evaluation of the tumor size according to the Response Evaluation Criteria In Solid Tumors (RECIST) [44]. Unfortunately, RECIST only reflects tumor size changes (which are often delayed, if they occur at all) and is unable to identify non-responders at an early time-point, when novel cytostatic biologic agents are employed [45]. A patient may be misclassified as a non-responder because the tumor size remains unchanged, or even increases in the early stages of treatment due to hemorrhage, necrosis, or edema, in spite of a decrease of the viable tumor. To add functional assessment, new methods that also reflect tumor perfusion have been introduced in the form of modified RECIST (mRECIST) criteria [46]. This has highlighted the need for alternative accurate and reproducible quantitative techniques to assess changes in tumor vascularity, a question which is not addressed satisfactorily by current standard diagnostic evaluation.

Clinical Applications

DCE-US quantification has been used to monitor changes induced by anti-angiogenic [47, 48] and anti-inflammatory [49, 50, 51, 52, 53] therapies, both as a potential marker of response and as a tool to enable dose optimization of therapy in individual patients [54]. Early clinical trials assessing tumor response in gastrointestinal stromal tumor (GIST) were based on the subjective and qualitative assessment of enhancement dynamics. Subsequent studies assessed response in renal cell carcinoma, hepatocellular carcinoma (HCC), breast cancer, pancreatic cancer, and colorectal metastases using semi-quantitative techniques [55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65]. Additional studies [50, 51, 56] used quantitative techniques to derive parameters related to the time course of contrast enhancement, in comparison to clinical endpoints such as Progression Free Survival (PFS) and Overall Survival (OS) following anti-angiogenic treatment. Techniques such as respiratory gating [59, 66, 67] and motion correction have been shown to improve the reproducibility of DCE-US measurements. A number of clinical trials have since evaluated DCE-US in therapy monitoring or intervention guidance, also demonstrating the potential of this technique in comparison to other imaging techniques such as DCE-MRI [53, 68, 69, 70, 71, 72, 73, 74, 75, 76], CT perfusion [77], or positron emission tomography [78]. Preliminary results have also been reported in children [79].

The number of clinical studies on DCE-US has increased since the initial publication in 2012, as well as the variety of technical approaches used to acquire and analyze contrast enhancement dynamics. The selection of these techniques may influence the reliability of reported results and possibly explain contrasting observations between studies. The following sections attempt to explain the available DCE-US techniques and parameters, with the aim of establishing a more standard approach to DCE-US examinations.

General considerations

Clinical DCE-US is usually performed with pure blood pool agents, such as SonoVue/Lumason [sulfur hexafluoride with a phospholipid shell, Bracco spa, Milan, Italy], or Definity [Octafluoropropane with a phospholipid shell, Lantheus Medical Imaging, Billerica MA, USA]. Quantitative contrast techniques can also be applied to agents, which are targeted to accumulate through specific biological interactions or to be extracted by a specific process (such as phagocytosis), but they require more complex multi-compartment kinetic models and are beyond the scope of this paper [43].

DCE-US can be performed using two different administration methods, an intravenous bolus injection or an infusion of UCA. The latter is followed by a disruption-replenishment technique and is much less commonly used for the assessment of tumor perfusion than the bolus injection.

The dual blood supply of the liver complicates blood flow quantification. After a bolus injection, the arterial blood supply is responsible for the initial enhancement of the normal parenchyma and of focal lesions, as the microbubbles arriving through the portal blood supply are delayed by 5 to 10 seconds. With the infusion technique, the replenishment reflects a combination of arterial and portal flow inputs.

After a bolus injection of UCA with wash-in/wash-out (bolus-transit) analysis, single-plane imaging at a low mechanical index (MI) is usually performed at about 10 frames per second for the duration of the enhancement. Frame rates that are too high should be avoided to prevent bubble destruction. Three-dimensional acquisition (corresponding to a volume) rather than a single plane would be preferable to overcome some limitations related to single plane analysis, but it is currently not feasible with the currently available commercial hardware (in terms of transducers and computing speed of available equipment). The average CEUS signal intensity within a region of interest (ROI) is calculated in linear units and is displayed as a function of time, i. e., a time-intensity curve (TIC), which describes the phases of progressive increase in enhancement of the contrast agent in the ROI (also termed wash-in) and the subsequent phase of slow decrease in contrast signal intensity (termed wash-out phase). Additional ROIs can be placed in a reference tissue for comparison purposes or in different areas of the lesion.

UCA administration

The approved doses are for bolus injection of SonoVue 2.4 mL for examinations of the macro- or microvasculature, and 2 mL SonoVue or Definity (10 µL/kg) in echocardiography. However, this dose may be reduced to 0.6–1.5 mL in most ultrasound systems, or increased up to the dose of two bolus injections (4.8 mL of SonoVue) under certain conditions depending on the sensitivity of the equipment, the transducer type and central frequency, the degree of vascularity, and the depth of the target lesion [80]. With more recent and sensitive equipment, the lower doses are adequate and should be preferred except when high-frequency transducers are

used. For example, the dose may be reduced to 1 mL when scanning the kidneys (and particularly in renal transplants), while it can be increased to 4.8 mL in the case of a superficial lesion using a high-frequency linear array or endoscopic transducers [81, 82, 83]. All microbubbles tend to go up in saline and should be shaken from time to time – or a pump should be used.

The bolus injection in general and also for quantitative DCE-US using SonoVue should be quick and be performed with a short angio-catheter typically 20G (never a smaller diameter than 22G to avoid disruption of the contrast microbubbles when they cross a too narrow catheter lumen), placed in an antecubital vein, without using a long extension line. A 3-way stop valve may be used at the end of the catheter to allow controlled access. In this case, it is preferable to connect the contrast syringe to the lock directly in line with the intravenous tract (not the perpendicular one) to avoid microbubble disruption that could occur when injecting contrast bolus against the stop valve tube wall. A saline flush (5 mL) should immediately follow to further sharpen the injected bolus and to limit the volume of UCA remaining in the angio-catheter and stop valve.

For infusion studies up to 2 vials (9.6 mL) have been infused at a rate of about 1 mL/min (or less) depending on the enhancement level required [84]. Slow infusion requires either a drip bag that is gently shaken from time to time or a pump that can be placed vertically or a specific rotating pump to continuously agitate the microbubbles [84]. Analysis should be performed during a steady UCA concentration in the blood. An acceptable steady state situation is usually achieved after about 2 minutes of infusion depending on the infusion rate. An initial faster injection rate can be used to achieve steady state earlier.

CEUS time intensity curve parameters

CEUS time intensity curve parameters have been summarized in the EFSUMB position paper describing the bolus-transit of the contrast microbubbles in the ROI [85, 86]. Time-related parameters can be differentiated from signal intensity-related parameters [3]. Several derived TIC parameters are purely descriptive/empirical. Reliability and potential sources of errors have been described [80].

Time-to-peak (TP), rise time (RT), mean transit time (MTT), peak intensity (PI), and area under the curve (AUC) have been proposed as primary parameters and all others are derived from those parameters [87]. In the EFSUMB position paper parameters such as time zero offset (T_0), time-to-peak (TP), wash-in time (WIT), wash-out time (WOT), mean transit time (MTT), full width half max (FWHM) are explained in detail [3]. Different from the other parameters, MTT can be calculated only in combination with a fitted mathematical model, while the other parameters are curve-descriptive parameters and thus can be derived also without a dedicated model. Since it is assumed that the signal intensity in DCE-US is proportional to the number of microbubbles (see below, linearized image data), and the microbubbles remain strictly intravascular, the TIC parameters are related to the vascularization of the analyzed region. Some signal-related parameters (peak intensity, area under the curve) are more correlated to the local blood volume of the region (~ mL), while other time-related parameters are more reflective of blood flow (TTP, WIT, AUC is also related to blood flow according to the Steward-Hamilton relationship). All time and intensity values should be calculated from a curve fitted to the linearized echo intensity values and not from image data.

Signal intensity-related parameters are given in arbitrary units [a.u.], with the most important being peak intensity (PI) and area under the curve (AUC). Both are described in detail in the already mentioned paper. The whole AUC describing the area under the curve may be divided into two components: the AUC of the wash-in phase up to peak intensity PI (WIAUC) and of the wash-out from peak intensity until the predefined time of end (WOAUC). The total AUC is the sum of WIAUC + WOAUC.

Other parameters include the wash-in rate (WIR) [a.u./s], which describes the slope of the TIC curve during wash-in [signal intensity/s]. The *maximum* slope of the TIC curve or the *mean* slope of a certain wash-in time interval (e. g., from 5 % to 95 % signal intensity) is used for this empirical parameter that is related to the blood flow. In a similar way, change during wash-out (WOR) can also be derived. In addition, combinations of the above parameters exist, in particular ratios between a signal intensity and a time-related parameter such as the wash-in perfusion index WIPI, which is the wash-in AUC divided by the wash-in time (WIAUC/WIT).

Refilling kinetics describe the replenishment of microbubbles during the infusion of UCA. UCA is first imaged without being disrupted at a low MI, then a few frames are acquired at a high MI (often at the highest available) causing bubble disruption in the image plane. Immediately thereafter, the MI is reverted to its low setting and the arrival of fresh microbubbles is imaged. Refilling kinetics are described by parameters that are different from those after bolus injection. T0 has an identical definition as for the TIC curves after bolus injection. TP, WIT, and MTT can also be calculated using a mathematical model that describes the refilling process. In contrast to bolus injection TIC curves, the maximum signal here is no longer reached at a peak but rather in the plateau phase. *Ip is the maximum signal* reached at the plateau (complete replenishment), often also called A, and is proportional to the local blood volume. The *rise of the replenishment curve [1/s],* often called B or β (based on the model by Wei *et al*., see below), is a parameter that is proportional to the local blood flow velocity. Since the replenishment curve usually has a sigmoidal shape, this parameter varies with time, and its concrete definition depends on the model used. In principle, A and B are parameters of the replenishment curves that are directly related to the blood volume and flow velocity (and its product is directly related to the blood flow, F = A*B). They can be extracted from the curves even without using a specific mathematical model that requires a closed form analytical expression, and thus they can easily be calculated and may be less prone to model-dependent limitations.

In 1998, Wei *et al*. [85] were the first to introduce the disruption-replenishment method and the development of the mono-exponential model. Krix *et al.*[88, 89, 90] used a similar approach as Wei *et al*. However, the modified formulas were no longer based on empiric assumptions and were based on a multi-vessel model incorporating differences in the acoustic field properties when using

high- and low-MI imaging. This model was found to be at least equivalent to the mono-exponential model, but it is nevertheless used much less frequently. Wei's model was improved by Arditi's model [91], which was subsequently further improved by Hudson *et al.*[92]. This model has 3 components that were not present in Wei's model: accounts for tissue perfusion through realistic microvascular geometry (Lognormal perfusion model), considers the ultrasound field properties of the destruction beam, and also considers the ultrasound imaging field. With the Arditi-Hudson model, it is possible to calculate the relative mean flow rate.

For repeated DCE-US exams, identical contrast protocols, DCE-US parameters, and analysis models have to be used in order to facilitate inter- or intra-patient comparison. Standardization and harmonization of software-based solutions and the various solutions integrated in the US platform are desirable but don't exist yet. For a detailed description of the equipment settings and patient-based factors, we refer to the published position papers [3]. Most studies focusing on AUC and wash-out recorded 3-minute loops [68]. In studies using infusion of UCA and the destruction-replenishment protocol, a shorter loop of the replenishment of the lesion or organ is sufficient (15–60 s) with the option to repeat it.

Clinical aspects of a DCE-US protocol

Choice of DCE-US parameters

A key question is which DCE-US technique and parameter should be used and evaluated in the various clinical settings. As described above, some parameters are more related to the blood volume (like the peak intensity Ip or the plateau A in replenishment kinetics) while others are more related to the dynamics of the blood supply, the blood flow (like the MTT). This is a first relevant aspect when choosing a certain DCE-US parameter. Furthermore, like with other imaging methods that analyze tissue vascularization (e. g., CT or MRI perfusion imaging in stroke) also a set of parameters and the identification of a potential mismatch between them may be useful to evaluate. In oncology treatment, monitoring or even outcome prediction are key aspects for use of DCE-US. This means parameters that could allow early assessment or prediction of treatment success or failure are the candidates of choice. In theory, changes in vascular dynamics (blood flow) would occur before a change in the vascular morphology (blood volume) becomes evident, but this has not yet been clearly demonstrated with DCE-US. Still, a widely used approach in research projects using DCE-US is to calculate more or less all feasible parameters and then to analyze if there is a correlation between these parameters and the specific clinical efficacy/outcome parameters. A few studies have suggested a certain parameter to be preferable in a specific setting (e. g., the MTT in bevacizumab therapy of metastases [53]) but a general broad consensus is lacking. AUC may be the most robust parameter in terms of technical errors.

DCE-US study design

Future studies should report DCE-US results in a more specific manner, related to certain parameters. Results should also be set in the context of the concrete tumor and treatment being assessed. "DCE-US for chemotherapy monitoring" may be a too broad and unspecific term. It should be clarified to which specific treatment or drug group study results are reported. In general, confirmatory studies are still needed to determine the crucial DCE-US parameters that should be focused on for the various clinical scenarios. It means an a priori hypothesis is to be proven in a prospective multicenter approach– such as "change of AUC tumor/AUC liver at time point x compared to baseline provides decisive information for therapy management with drug *xy*". So, a very narrow study hypothesis focusing on concrete parameters, time points, etc. and using a valuable clinical endpoint should be applied. Several studies so far have been explorative and have only used another biomarker for comparison such as perfusion in MRI or microvessel density in pathology.

Here, a comparison between classic early RECIST and DCE-US results is per se of no or low additional clinical value. Studies should rather focus on the potential additive value of DCE-US compared to standard diagnostics, i. e., on the predictive value of the method at early time points. The use of long-term outcome data as the standard of reference should be preferred to demonstrate whether DCE-US performs better at follow-up compared to RECIST.

DCE-US exam time points

This is related to the question at which clinical time point(s) a DCE-US examination should be performed. Treatment monitoring requires follow-up examinations while predictive messages or data for guidance of interventions can be derived from a single, early exam. Clinical trials using DCE-US in monitoring have often focused on standard time points, i. e., before the start of treatment and follow-up exams performed at standard time points in parallel to established imaging (e. g., for RECIST). Early time points for follow-up sometimes have been added, in particular a DCE-US exam after a first cycle of chemotherapy. Currently, further studies are still needed to determine the optimum monitoring regime for a specific treatment. Not only the duration after the general start of treatment can be relevant, but also the duration after a certain cycle of chemotherapy can have a considerable impact. Anti-angiogenic effects may be observable already within a short period of time, maybe the optimum only within a specific period of time after administration. DCE-US should clearly report how the used exam time points have been chosen and further studies can increase knowledge for optimization of monitoring schemes.

How to perform DCE-US, how to interpret the results, technical advice

To optimize the machine settings for DCE-CEUS, the following issues are important. One single focus position should be set in a deeper region of the scanning plane, which must include by large all the regions of interest. The lowest but still reliable mechanical index (MI) should be used to avoid any unnecessary bubble disruption. The most convenient MI value varies depending on the specific equipment. The receive gain should be set so that it is usually aligned in the middle position. The persistence mode

should be turned off and the dynamic range should be kept tendentially high despite the fact that these two adjustments may not provide the best ultrasound images.

The conditions of the patient and surrounding factors (including posture, resting time, heart rate, blood pressure) and also of the scanning plane (acoustic window, probe position) at each acquisition, to help explain discrepancies in unexpected findings taken at different sessions during follow-up should be standardized and recorded. For adequate reproducibility, the follow-up examinations require the scanning plane to be exactly the same. This is often very difficult to achieve, even for expert users. A clear description of the probe position for examining the lesion, with landmarks in relation to the skin surface and documentation of representative anatomical structures, e. g., liver segment(s), major vessels, as well as the CEUS acquisition parameters, such as the depth of the lesion, mechanical index, etc., are essential to ensure standardization of these subsequent studies. It is important to keep all imaging (machine) parameters unchanged after the baseline scan to allow the comparison of the effects of therapy in subsequent scans.

Find a tumor in conventional B-mode and choose a tumor plane to study. Inject the appropriate dosage of microbubble contrast agents and scan in contrast mode (side-by-side). In order to keep the probe stationary, be aware of and compensate for any motion. Some examiners have also used an articulated arm to stay on the same plane. Scan continuously for 2 up to 5 minutes (depending on the clinical application) avoiding bubble destruction. Commercially available software (e. g., Vuebox) also allows the merging of smaller videos into one video, which can be helpful in the case of motion but also to reduce bubble destruction. Save the DICOM video loop in a format that allows data linearization. If more than one TIC curve may be recorded, then rotate the probe to select a different tumor plane (to evaluate tumor heterogeneity) and repeat the steps above for both infusion and replenishment.

The data analysis involves the use of a software package that allows forming of the TIC from linearized data from ROIs in the lesion and one in the normal parenchyma. One ROI should cover the whole tumor, and the placement of optional additional ROI(s) should follow representative areas of the "whole tumor" guided by highly vascularized parts of the tumor. For early relapse prediction, focusing on highly vascularized ROIs may be useful. In partially necrotic tumors, this guidance can make an important difference. For some of the mentioned recommendations, no consensus has been reached so far.

Next, a curve is fitted to the TIC data and the important perfusion parameters (rise time RT, mean transit time MTT, peak intensity PI, and area under the curve AUC) are extracted. Interpretation of the results involves statistically correlating the perfusion parameters with physiological data and clinical outcomes.

Further technical and methodological aspects

Technical considerations also contribute to the choice of an optimum DCE-US parameter. Reproducibility is an important factor, and DCE-US exams can be influenced by various aspects. Thus, the most robust parameters can be preferable. Time-related parameters (rise time) are robust since they do not depend on the acoustic signal level – if the bolus arrival time is subtracted to avoid circulation time dependencies. Integrals are per se more robust than single values, thus the AUC or also parameters based on a mathematical integral and a closed form analytical expression (MTT) can be beneficial in the clinical routine, but this has been a topic of controversial discussion between the authors. However, the quality of the fit must be recorded to avoid misinterpretation. Even then, all parameters related to the CEUS signal intensity can crucially be influenced and biased by the various acoustic and patient conditions which may drastically limit inter- or longitudinal intra-patient comparison. This is mainly related to signal-related parameters. Normalization is the key to reducing this variability. Instead of using parameters of a single ROI (in oncology usually this is the tumor), values obtained from this ROI should be normalized, usually placing a second ROI in normal tissue adjacent to the relevant tissue (for instance the liver) and calculating the ratio of the parameters in these ROIs. Such normalized parameters are less prone to external bias. Time-related parameters are less influenced but also these parameters of a tumor can be compared with the surrounding tissue, e. g., as the difference in the rise time in the tumor compared to the liver.

The choice of the US plane may be affected by the visibility of representative tissue. The second ROI should be placed at the same depth. If no normal (healthy) parenchyma is present, other normal organs visible in the US plane could be used as an exception. Due to signal linearization, large vessels should not be used as the standard ROI for comparison. The focus should be positioned just at the level of the target lesion for most ultrasound scanners. Deeper focal zones might be used to achieve a more uniform acoustic field, which improves sensitivity to the agents and lessens the risk of bubble disruption. Detailed general technical recommendations have been published elsewhere in a consensus paper [93].

Perspectives

Modern oncologic therapies not only aim at a decrease in tumor size but may also focus on a "return to normal" situation, i. e., a tumor then may still have a high but relatively normal blood volume or perfusion. Thus, even more sophisticated DCE-US parameters beyond those related to blood volume or perfusion could be needed to describe DCE-US patterns correlated with the vascular architecture. Existing models are able to derive such additional information, but they are not used in clinical practice. Finally, the described DCE-US parameters provide a temporal analysis of DCE-US exams, not a spatial analysis. Vessel architecture analysis, however, also requires a spatial component. Placing more than one ROI in a tumor, e. g., in the periphery and the center, is the simplest approach to add a spatial analysis. When color-coded parameter maps are generated with suitable software, more complex approaches are feasible, up to a pixel-wise comparison and correlation of DCE-US parameters. A simple spatial approach is also to use the size of the colored area in a tumor as an additional parameter – it is not pure DCE-US but DCE-US is used here to create such spatial parameters. For instance, the size of the AUC above a certain threshold/size of the whole tumor can reflect the vascularization – similar to "% of vascularized tumor". A combination of both a spatiotemporal analysis and the use of 3 D-US for

DCE-US may provide a more complete description of the UCA transport process and better characterization of perfusion, contrast dispersion, and vascular architecture [43]. In brain perfusion studies using MRI, such parameters are relevant – e. g., to identify a mismatch between perfusion parameters and to see if there is viable tissue at risk that justifies treatment after stroke. In regard to 3 D DCE-US, CEUS techniques are limited and publications are lacking [94], and these topics are beyond the scope of this document. Variability studies using phantoms and models across multiple scanners and quantification software have been described in detail; refer to [80, 87].

Although promising, all studies identified so far on AI in CEUS are single-center, retrospective studies, or studies on limited, selected case series using different algorithms for machine learning and with various clinical aims, even if characterization of liver lesions is the most frequent. Most often the algorithms are run in post-processing, making them less useful in a clinical workflow. There is a need to perform prospective, multi-center studies with clinically useful endpoints, preferably using open-access software in order to find the place for AI in the evaluation of CEUS cine loops.

Open questions

TIC curve analysis of CEUS bolus injections provides several parameters that reflect local blood flow. None of the parameters alone represent clear-cut tissue characterizing abilities, although differences are observed for e. g., neoplastic and non-neoplastic tissue [37, 95, 96, 97, 98, 99]. The ability to combine several parameters simultaneously using AI may provide improved characterization, but this must be shown in prospective multi-center studies using standardized technology. The dynamic contrast assessment methods need to be integrated in the clinical workflow, not requiring too much time, and the results should provide information influencing the clinical management of patients.

Concluding remarks

Results of recent monocentric and multicentric clinical trials propose that quantitative DCE-US may be useful in oncology, in particular in the assessment of response to targeted therapies beyond classic RECIST assessment. The current article provides general information about the technique and parameters utilized in DCE-US quantification and recommendations on its use to provide a standardized approach, which may improve clinical management.

Statements

STATEMENT 1

Compared to a purely subjective comparison of the phase-specific enhancement of different tissues or of the same tissue under different pathological or therapeutic conditions, DCE-US allows a more objective assessment when used in a standardized way.

STATEMENT 2

Using only tumor diameter changes (i. e., RECIST) is a suboptimal method for tumor response assessment. Treatment monitoring assessment of vascularization/perfusion adds relevant information both in the early and later phases after initiation of pharmacological treatments.

STATEMENT 3

Further research is recommended to investigate the potential of DCE-US to noninvasively improve the differential diagnosis of focal lesions in parenchymal organs, to graduate the biological aggressiveness of various malignant tumors, and to predict their outcome, as well as to record the temporal dynamics of pathological processes in parenchymal organs associated with changes in perfusion characteristics.

STATEMENT 4

DCE-US provides quantitative information about local blood flow and can be carried out with two main DCE-US modalities, which provide different information and parameters: the bolus technique and the infusion technique (using the disruption-replenishment method).

STATEMENT 5

The bolus technique quantifies the entire course of contrast kinetics, from wash-in to wash-out. The analysis is carried out along one single plane for each injection and a cineloop of at least one minute in duration is recommended. The disruption-replenishment method is carried out at a steady-state high signal enhancement level. The analysis requires a shorter cineloop (usually 10–25 seconds), so that multiple planes can be assessed. Parameters and information obtained with the two methods differ from each other.

STATEMENT 6

Relative quantification of perfusion using a reference area at the same depth should be preferred to absolute evaluation of contrast enhancement.

STATEMENT 7

In order to optimize machine settings for DCE-CEUS, the following recommendations are important: a) use a single focus

position to be set in a deeper region of the scanning plane that must include all regions of interest; b) use a low mechanical index (MI); c) set the receive gain high with TGC usually aligned in the middle position; d) turn off the persistence mode and keep the dynamic range tendentially high despite the fact that these two adjustments may not provide the best B-mode ultrasound images.

STATEMENT 8

The MI should be set as low as possible, with the goal of avoiding any unnecessary bubble disruption. The most convenient MI value varies depending on the specific equipment and the contrast agent being used.

STATEMENT 9

To assess tumor response in a patient, the same machine settings should be used for consecutive DCE-US examinations as for the baseline examination. It is recommended to keep a detailed record of patient conditions and surrounding factors (including posture, resting time, heart rate, blood pressure) and also the scanning plane (acoustic window, probe position) for each acquisition, to help explain discrepancies in unexpected findings taken in different sessions during follow-up.

STATEMENT 10

Suitable planning and choice of a representative imaging plane is crucial to avoid respiratory motion of the ROI which is a major source of error in the quantification of DCE-US. Especially out-of-plane motion cannot be corrected, and out-of-plane acquisitions must be excluded from the DCE-US analysis, which is a time-consuming and demanding process.

STATEMENT 11

Quantification software may be embedded in ultrasound equipment or may be work off-line on separate hardware. It is necessary to perform calculations on linearized data to maintain the linear relationship between microbubble concentration and signal intensity.

Acknowledgement

The authors thank Dr. Kim Nylund, Prof. Mike Averkiou and Prof. Peter N Burns for important input and advice.

Conflict of Interest

All other authors have received lecture honoraria from Bracco and from companies selling ultrasound systems. Martin Krix is employee of Bracco.

References

[1] Albrecht T, Blomley M, Bolondi L et al. Guidelines for the use of contrast agents in ultrasound. Ultraschall in der Medizin (Stuttgart, Germany : 1980) 2004; 25: 249–256. doi:10.1055/s-2004-813245

[2] Claudon M, Cosgrove D, Albrecht T et al. Guidelines and good clinical practice recommendations for contrast enhanced ultrasound (CEUS) – update 2008. Ultraschall in der Medizin (Stuttgart, Germany : 1980) 2008; 29: 28–44. doi:10.1055/s-2007-963785

[3] Dietrich CF, Averkiou MA, Correas JM et al. An EFSUMB introduction into Dynamic Contrast-Enhanced Ultrasound (DCE-US) for quantification of tumour perfusion. Ultraschall in der Medizin (Stuttgart, Germany : 1980) 2012; 33: 344–351. doi:10.1055/s-0032-1313026

[4] Claudon M, Dietrich CF, Choi BI et al. Guidelines and good clinical practice recommendations for Contrast Enhanced Ultrasound (CEUS) in the liver – update 2012: A WFUMB-EFSUMB initiative in cooperation with representatives of AFSUMB, AIUM, ASUM, FLAUS and ICUS. Ultrasound in medicine & biology 2013; 39: 187–210. doi:10.1016/j.ultrasmedbio.2012.09.002

[5] Claudon M, Dietrich CF, Choi BI et al. Guidelines and good clinical practice recommendations for contrast enhanced ultrasound (CEUS) in the liver–update 2012: a WFUMB-EFSUMB initiative in cooperation with representatives of AFSUMB, AIUM, ASUM, FLAUS and ICUS. Ultraschall in der Medizin (Stuttgart, Germany : 1980) 2013; 34: 11–29. doi:10.1055/s-0032-1325499

[6] Dietrich CF, Nolsoe CP, Barr RG et al. Guidelines and Good Clinical Practice Recommendations for Contrast-Enhanced Ultrasound (CEUS) in the Liver-Update 2020 WFUMB in Cooperation with EFSUMB, AFSUMB, AIUM, and FLAUS. Ultrasound in medicine & biology 2020; 46: 2579–2604. doi:10.1016/j.ultrasmedbio.2020.04.030

[7] Dietrich CF, Nolsoe CP, Barr RG et al. Guidelines and Good Clinical Practice Recommendations for Contrast Enhanced Ultrasound (CEUS) in the Liver – Update 2020 – WFUMB in Cooperation with EFSUMB, AFSUMB, AIUM, and FLAUS. Ultraschall in der Medizin (Stuttgart, Germany : 1980) 2020; 41: 562–585. doi:10.1055/a-1177-0530

[8] Wustner M, Radzina M, Calliada F et al. Professional Standards in Medical Ultrasound – EFSUMB Position Paper (Long Version) – General Aspects. Ultraschall in der Medizin (Stuttgart, Germany : 1980) 2022; 43: e36–e48. doi:10.1055/a-1857-4435

[9] Jenssen C, Gilja OH, Serra AL et al. European Federation of Societies for Ultrasound in Medicine and Biology (EFSUMB) Policy Document Development Strategy – Clinical Practice Guidelines, Position Statements and Technological Reviews. Ultrasound Int Open 2019; 5: E2–E10. doi:10.1055/a-0770-3965

[10] Lupusoru R, Sporea I, Ratiu I et al. Contrast-Enhanced Ultrasonography with Arrival Time Parametric Imaging as a Non-Invasive Diagnostic Tool for Liver Cirrhosis. Diagnostics (Basel, Switzerland) 2022; 12. doi:10.3390/diagnostics12123013

[11] Zocco MA, Cintoni M, Ainora ME et al. Noninvasive Evaluation of Clinically Significant Portal Hypertension in Patients with Liver Cirrhosis: The Role of Contrast-Enhanced Ultrasound Perfusion Imaging and Elastography. Ultraschall in der Medizin (Stuttgart, Germany : 1980) 2022. doi:10.1055/a-1933-2847

[12] Wakui N, Nagai H, Ogino Y et al. Hepatic arterialization can predict the development of collateral veins in patients with HCV-related liver disease. J Ultrasound 2018; 21: 301–308. doi:10.1007/s40477-018-0323-4

[13] Kim G, Shim KY, Baik SK. Diagnostic Accuracy of Hepatic Vein Arrival Time Performed with Contrast-Enhanced Ultrasonography for Cirrhosis:

A Systematic Review and Meta-Analysis. Gut Liver 2017; 11: 93–101. doi:10.5009/gnl16031

[14] Kiyono S, Maruyama H, Kobayashi K et al. Non-Invasive Diagnosis of Portal Hypertensive Gastropathy: Quantitative Analysis of Microbubble-Induced Stomach Wall Enhancement. Ultrasound in medicine & biology 2016; 42: 1792–1799. doi:10.1016/j.ultrasmedbio.2016.03.011

[15] Nasr P, Hilliges A, Thorelius L et al. Contrast-enhanced ultrasonography could be a non-invasive method for differentiating none or mild from severe fibrosis in patients with biopsy proven non-alcoholic fatty liver disease. Scand J Gastroenterol 2016; 51: 1126–1132. doi:10.3109/00365521.2016.1172336

[16] Cavorsi K, Prabhakar P, Kirby C. Acute Pyelonephritis. Ultrasound Quarterly 2010; 26: 103–105. doi:10.1097/RUQ.0b013e3181dc7d0b

[17] Ma F, Cang Y, Zhao B et al. Contrast-enhanced ultrasound with SonoVue could accurately assess the renal microvascular perfusion in diabetic kidney damage. Nephrol Dial Transplant 2012; 27: 2891–2898. doi:10.1093/ndt/gfr789

[18] Srivastava A, Sridharan A, Walmer RW et al. Association of Contrast-Enhanced Ultrasound-Derived Kidney Cortical Microvascular Perfusion with Kidney Function. Kidney360 2022; 3: 647–656. doi:10.34067/KID.0005452021

[19] Wang L, Wu J, Cheng JF et al. Diagnostic value of quantitative contrast-enhanced ultrasound (CEUS) for early detection of renal hyperperfusion in diabetic kidney disease. J Nephrol 2015; 28: 669–678. doi:10.1007/s40620-015-0183-3

[20] Kim DG, Lee JY, Ahn JH et al. Quantitative ultrasound for non-invasive evaluation of subclinical rejection in renal transplantation. Eur Radiol 2022. doi:10.1007/s00330-022-09260-x

[21] Zhao P, Li N, Lin L et al. Correlation between serum cystatin C level and renal microvascular perfusion assessed by contrast-enhanced ultrasound in patients with diabetic kidney disease. Ren Fail 2022; 44: 1732–1740. doi:10.1080/0886022X.2022.2134026

[22] Zhang W, Yi H, Cai B et al. Feasibility of contrast-enhanced ultrasonography (CEUS) in evaluating renal microvascular perfusion in pediatric patients. BMC Med Imaging 2022; 22: 194. doi:10.1186/s12880-022-00925-z

[23] He L, Li Z, Zhang Q et al. Evaluation of renal microperfusion in hyperuricemic nephropathy by contrast-enhanced ultrasound imaging. Dis Model Mech 2022; 15. doi:10.1242/dmm.049382

[24] Garessus J, Brito W, Loncle N et al. Cortical perfusion as assessed with contrast-enhanced ultrasound is lower in patients with chronic kidney disease than in healthy subjects but increases under low salt conditions. Nephrol Dial Transplant 2022; 37: 705–712. doi:10.1093/ndt/gfab001

[25] Puca P, DelVecchio LE, Ainora ME et al. Role of Multiparametric Intestinal Ultrasound in the Evaluation of Response to Biologic Therapy in Adults with Crohn's Disease. Diagnostics (Basel, Switzerland) 2022; 12. doi:10.3390/diagnostics12081991

[26] Ding SS, Liu C, Zhang YF et al. Contrast-enhanced ultrasound in the assessment of Crohn's disease activity: comparison with computed tomography enterography. Radiol Med 2022; 127: 1068–1078. doi:10.1007/s11547-022-01535-z

[27] Ponorac S, Dahmane Gosnak R, Urlep D et al. Diagnostic Value of Quantitative Contrast-Enhanced Ultrasound in Comparison to Endoscopy in Children With Crohn's Disease. J Ultrasound Med 2023; 42: 193–200. doi:10.1002/jum.16044

[28] Freitas M, de Castro FD, Macedo Silva V et al. Ultrasonographic scores for ileal Crohn's disease assessment: Better, worse or the same as contrast-enhanced ultrasound? BMC Gastroenterol 2022; 22: 252. doi:10.1186/s12876-022-02326-6

[29] Servais L, Boschetti G, Meunier C et al. Intestinal Conventional Ultrasonography, Contrast-Enhanced Ultrasonography and Magnetic Resonance Enterography in Assessment of Crohn's Disease Activity: A Comparison with Surgical Histopathology Analysis. Dig Dis Sci 2022; 67: 2492–2502. doi:10.1007/s10620-021-07074-3

[30] Laterza L, Ainora ME, Garcovich M et al. Bowel contrast-enhanced ultrasound perfusion imaging in the evaluation of Crohn's disease patients undergoing anti-TNFalpha therapy. Dig Liver Dis 2021; 53: 729–737. doi:10.1016/j.dld.2020.08.005

[31] Zezos P, Zittan E, Islam S et al. Associations between quantitative evaluation of bowel wall microvascular flow by contrast-enhanced ultrasound and indices of disease activity in Crohn's disease patients using both bolus and infusion techniques. J Clin Ultrasound 2019; 47: 453–460. doi:10.1002/jcu.22763

[32] Ripolles T, Martinez-Perez MJ, Paredes JM et al. The Role of Intravenous Contrast Agent in the Sonographic Assessment of Crohn's Disease Activity: Is Contrast Agent Injection Necessary? J Crohns Colitis 2019; 13: 585–592. doi:10.1093/ecco-jcc/jjy204

[33] Nylund K, Saevik F, Leh S et al. Interobserver Analysis of CEUS-Derived Perfusion in Fibrotic and Inflammatory Crohn's Disease. Ultraschall in der Medizin (Stuttgart, Germany : 1980) 2019; 40: 76–84. doi:10.1055/s-0044-100492

[34] Goertz RS, Klett D, Wildner D et al. Quantitative contrast-enhanced ultrasound for monitoring vedolizumab therapy in inflammatory bowel disease patients: a pilot study. Acta Radiol 2018; 59: 1149–1156. doi:10.1177/0284185117752032

[35] Liu H, Cao H, Chen L et al. The quantitative evaluation of contrast-enhanced ultrasound in the differentiation of small renal cell carcinoma subtypes and angiomyolipoma. Quant Imaging Med Surg 2022; 12: 106–118. doi:10.21037/qims-21-248

[36] Liang RX, Wang H, Zhang HP et al. The value of real-time contrast-enhanced ultrasound combined with CT enhancement in the differentiation of subtypes of renal cell carcinoma. Urol Oncol 2021; 39: 837e19–837e28. doi:10.1016/j.urolonc.2021.09.004

[37] Dong Y, Qiu Y, Yang D et al. Potenzial application of dynamic contrast enhanced ultrasound in predicting microvascular invasion of hepatocellular carcinoma. Clin Hemorheol Microcirc 2021; 77: 461–469. doi:10.3233/CH-201085

[38] Liu Z, Li C. Correlation of lymph node metastasis with contrast-enhanced ultrasound features, microvessel density and microvessel area in patients with papillary thyroid carcinoma. Clin Hemorheol Microcirc 2022; 82: 361–370. doi:10.3233/CH-221545

[39] Xuan Z, Wu N, Li C et al. Application of contrast-enhanced ultrasound in the pathological grading and prognosis prediction of hepatocellular carcinoma. Transl Cancer Res 2021; 10: 4106–4115. doi:10.21037/tcr-21-1264

[40] Li X, Han X, Li L et al. Dynamic Contrast-Enhanced Ultrasonography with Sonazoid for Diagnosis of Microvascular Invasion in Hepatocellular Carcinoma. Ultrasound in medicine & biology 2022; 48: 575–581. doi:10.1016/j.ultrasmedbio.2021.11.005

[41] Feng Y, Qin XC, Luo Y et al. Efficacy of contrast-enhanced ultrasound washout rate in predicting hepatocellular carcinoma differentiation. Ultrasound in medicine & biology 2015; 41: 1553–1560. doi:10.1016/j.ultrasmedbio.2015.01.026

[42] El Kaffas A, Hoogi A, Zhou J et al. Spatial Characterization of Tumor Perfusion Properties from 3D DCE-US Perfusion Maps are Early Predictors of Cancer Treatment Response. Sci Rep 2020; 10: 6996. doi:10.1038/s41598-020-63810-1

[43] Turco S, Frinking P, Wildeboer R et al. Contrast-Enhanced Ultrasound Quantification: From Kinetic Modeling to Machine Learning. Ultrasound in medicine & biology 2020; 46: 518–543. doi:10.1016/j.ultrasmedbio.2019.11.008

[44] Eisenhauer EA, Therasse P, Bogaerts J et al. New response evaluation criteria in solid tumours: revised RECIST guideline (version 1.1). European journal of cancer (Oxford, England : 1990) 2009; 45: 228–247. doi:10.1016/j.ejca.2008.10.026

[45] Zhu AX, Holalkere NS, Muzikansky A et al. Early antiangiogenic activity of bevacizumab evaluated by computed tomography perfusion scan in patients with advanced hepatocellular carcinoma. The oncologist 2008; 13: 120–125. doi:10.1634/theoncologist.2007-0174

[46] Lencioni R, Llovet JM. Modified RECIST (mRECIST) assessment for hepatocellular carcinoma. Seminars in liver disease 2010; 30: 52–60. doi:10.1055/s-0030-1247132

[47] Leen E, Averkiou M, Arditi M et al. Dynamic contrast enhanced ultrasound assessment of the vascular effects of novel therapeutics in early stage trials. Eur Radiol 2012; 22: 1442–1450. doi:10.1007/s00330-011-2373-2

[48] Hudson JM, Williams R, Tremblay-Darveau C et al. Dynamic contrast enhanced ultrasound for therapy monitoring. Eur J Radiol 2015; 84: 1650–1657. doi:10.1016/j.ejrad.2015.05.013

[49] Saevik F, Nylund K, Hausken T et al. Bowel perfusion measured with dynamic contrast-enhanced ultrasound predicts treatment outcome in patients with Crohn's disease. Inflammatory bowel diseases 2014; 20: 2029–2037. doi:10.1097/MIB.0000000000000159

[50] Socaciu M, Ciobanu L, Diaconu B et al. Non-Invasive Assessment of Inflammation and Treatment Response in Patients with Crohn's Disease and Ulcerative Colitis using Contrast-Enhanced Ultrasonography Quantification. Journal of gastrointestinal and liver diseases : JGLD 2015; 24: 457–465. doi:10.15403/jgld.2014.1121.244.chr

[51] De Franco A, Di Veronica A, Armuzzi A et al. Ileal Crohn disease: mural microvascularity quantified with contrast-enhanced US correlates with disease activity. Radiology 2012; 262: 680–688. doi:10.1148/radiol.11110440

[52] Medellin-Kowalewski A, Wilkens R, Wilson A et al. Quantitative Contrast-Enhanced Ultrasound Parameters in Crohn Disease: Their Role in Disease Activity Determination With Ultrasound. AJR Am J Roentgenol 2016; 206: 64–73. doi:10.2214/AJR.15.14506

[53] Lassau N, Coiffier B, Kind M et al. Selection of an early biomarker for vascular normalization using dynamic contrast-enhanced ultrasonography to predict outcomes of metastatic patients treated with bevacizumab. Annals of oncology : official journal of the European Society for Medical Oncology 2016; 27: 1922–1928. doi:10.1093/annonc/mdw280

[54] Bjarnason GA, Khalil B, Hudson JM et al. Outcomes in patients with metastatic renal cell cancer treated with individualized sunitinib therapy: correlation with dynamic microbubble ultrasound data and review of the literature. Urol Oncol 2014; 32: 480–487. doi:10.1016/j.urolonc.2013.10.004

[55] Lassau N, Koscielny S, Albiges L et al. Metastatic renal cell carcinoma treated with sunitinib: early evaluation of treatment response using dynamic contrast-enhanced ultrasonography. Clinical cancer research : an official journal of the American Association for Cancer Research 2010; 16: 1216–1225. doi:10.1158/1078-0432.ccr-09-2175

[56] Lassau N, Chami L, Koscielny S et al. Quantitative functional imaging by dynamic contrast enhanced ultrasonography (DCE-US) in GIST patients treated with masatinib. Investigational new drugs 2012; 30: 765–771. doi:10.1007/s10637-010-9592-2

[57] Lassau N, Koscielny S, Chami L et al. Advanced hepatocellular carcinoma: early evaluation of response to bevacizumab therapy at dynamic contrast-enhanced US with quantification–preliminary results. Radiology 2011; 258: 291–300. doi:10.1148/radiol.10091870

[58] Williams R, Hudson JM, Lloyd BA et al. Dynamic microbubble contrast-enhanced US to measure tumor response to targeted therapy: a proposed clinical protocol with results from renal cell carcinoma patients receiving antiangiogenic therapy. Radiology 2011; 260: 581–590. doi:10.1148/radiol.11101893

[59] Averkiou M, Lampaskis M, Kyriakopoulou K et al. Quantification of tumor microvascularity with respiratory gated contrast enhanced ultrasound for monitoring therapy. Ultrasound in medicine & biology 2010; 36: 68–77. doi:10.1016/j.ultrasmedbio.2009.07.005

[60] Trenker C, Kumpel J, Michel C et al. Assessment of Early Therapy Response of Non-Hodgkin's and Hodgkin's Lymphoma Using B-Mode Ultrasound and Dynamic Contrast-Enhanced Ultrasound. J Ultrasound Med 2022; 41: 2033–2040. doi:10.1002/jum.15887

[61] Huang Z, Zhu RH, Xin JY et al. HCC treated with immune checkpoint inhibitors: a hyper-enhanced rim on Sonazoid-CEUS Kupffer phase images is a predictor of tumor response. Eur Radiol 2022. doi:10.1007/s00330-022-09339-5

[62] Takada H, Yamashita K, Osawa L et al. Prediction of Therapeutic Response Using Contrast-Enhanced Ultrasound in Japanese Patients Treated with Atezolizumab and Bevacizumab for Unresectable Hepatocellular Carcinoma. Oncology 2022. doi:10.1159/000527752

[63] Guo J, Wang BH, He M et al. Contrast-enhanced ultrasonography for early prediction of response of neoadjuvant chemotherapy in breast cancer. Front Oncol 2022; 12: 1026647. doi:10.3389/fonc.2022.1026647

[64] Lu XY, Guo X, Zhang Q et al. Early Assessment of Chemoradiotherapy Response for Locally Advanced Pancreatic Ductal Adenocarcinoma by Dynamic Contrast-Enhanced Ultrasound. Diagnostics (Basel, Switzerland) 2022; 12. doi:10.3390/diagnostics12112662

[65] Zhou B, Lian Q, Jin C et al. Human clinical trial using diagnostic ultrasound and microbubbles to enhance neoadjuvant chemotherapy in HER2- negative breast cancer. Front Oncol 2022; 12: 992774. doi:10.3389/fonc.2022.992774

[66] Christofides D, Leen EL, Averkiou MA. Improvement of the accuracy of liver lesion DCEUS quantification with the use of automatic respiratory gating. Eur Radiol 2016; 26: 417–424. doi:10.1007/s00330-015-3797-x

[67] Christofides D, Leen E, Averkiou M. Automatic respiratory gating for contrast ultrasound evaluation of liver lesions. IEEE transactions on ultrasonics, ferroelectrics, and frequency control 2014; 61: 25–32. doi:10.1109/TUFFC.2014.6689773

[68] Lassau N, Chami L, Chebil M et al. Dynamic contrast-enhanced ultrasonography (DCE-US) and anti-angiogenic treatments. Discovery medicine 2011; 11: 18–24

[69] Green RW, Epstein E. Can dynamic contrast-enhanced ultrasound (DCE-US) improve diagnostic performance in endometrial cancer staging? A proof of concept. Ultrasound Obstet Gynecol 2019. doi:10.1002/uog.21885

[70] Yang Z, Kang M, Zhu S et al. Clinical evaluation of vascular normalization induced by recombinant human endostatin in nasopharyngeal carcinoma via dynamic contrast-enhanced ultrasonography. OncoTargets and therapy 2018; 11: 7909–7917. doi:10.2147/ott.s181842

[71] Zhan Y, Zhou F, Yu X et al. Quantitative dynamic contrast-enhanced ultrasound may help predict the outcome of hepatocellular carcinoma after microwave ablation. International journal of hyperthermia : the official journal of European Society for Hyperthermic Oncology, North American Hyperthermia Group 2019; 35: 105–111. doi:10.1080/02656736.2018.1483533

[72] Kim Y, Kim SH, Song BJ et al. Early Prediction of Response to Neoadjuvant Chemotherapy Using Dynamic Contrast-Enhanced MRI and Ultrasound in Breast Cancer. Korean journal of radiology 2018; 19: 682–691. doi:10.3348/kjr.2018.19.4.682

[73] Hudson JM, Bailey C, Atri M et al. The prognostic and predictive value of vascular response parameters measured by dynamic contrast-enhanced-CT, -MRI and -US in patients with metastatic renal cell carcinoma receiving sunitinib. Eur Radiol 2018; 28: 2281–2290. doi:10.1007/s00330-017-5220-2

[74] Mogensen MB, Hansen ML, Henriksen BM et al. Dynamic Contrast-Enhanced Ultrasound of Colorectal Liver Metastases as an Imaging Modality for Early Response Prediction to Chemotherapy. Diagnostics (Basel, Switzerland) 2017; 7: 10. doi:10.3390/diagnostics7020035

[75] Postema AW, Frinking PJ, Smeenge M et al. Dynamic contrast-enhanced ultrasound parametric imaging for the detection of prostate cancer. BJU international 2016; 117: 598–603. doi:10.1111/bju.13116

[76] Wilkens R, Peters DA, Nielsen AH et al. Dynamic Contrast-Enhanced Magnetic Resonance Enterography and Dynamic Contrast-Enhanced Ultrasonography in Crohn's Disease: An Observational Comparison Study. Ultrasound Int Open 2017; 3: E13–E24. doi:10.1055/s-0042-123841

[77] Frampas E, Lassau N, Zappa M et al. Advanced Hepatocellular Carcinoma: early evaluation of response to targeted therapy and prognostic value of Perfusion CT and Dynamic Contrast Enhanced-Ultrasound. Preliminary results. Eur J Radiol 2013; 82: e205–211. doi:10.1016/j.ejrad.2012.12.004

[78] Niermann KJ, Fleischer AC, Huamani J et al. Measuring tumor perfusion in control and treated murine tumors: correlation of microbubble contrast-enhanced sonography to dynamic contrast-enhanced magnetic resonance imaging and fluorodeoxyglucose positron emission tomography. J Ultrasound Med 2007; 26: 749–756. doi:10.7863/jum.2007.26.6.749

[79] McCarville MB, Coleman JL, Guo J et al. Use of Quantitative Dynamic Contrast-Enhanced Ultrasound to Assess Response to Antiangiogenic Therapy in Children and Adolescents With Solid Malignancies: A Pilot Study. AJR Am J Roentgenol 2016; 206: 933–939. doi:10.2214/AJR.15.15789

[80] Ignee A, Jedrejczyk M, Schuessler G et al. Quantitative contrast enhanced ultrasound of the liver for time intensity curves-Reliability and potential sources of errors. EurJRadiol 2010; 73: 153–158. doi:10.1016/j.ejrad.2008.10.016

[81] Piscaglia F, Nolsoe C, Dietrich CF et al. The EFSUMB Guidelines and Recommendations on the Clinical Practice of Contrast Enhanced Ultrasound (CEUS): update 2011 on non-hepatic applications. Ultraschall in der Medizin (Stuttgart, Germany : 1980) 2012; 33: 33–59. doi:10.1055/s-0031-1281676

[82] Dietrich CF, Ignee A, Frey H. Contrast-enhanced endoscopic ultrasound with low mechanical index: a new technique. Zeitschrift fur Gastroenterologie 2005; 43: 1219–1223. doi:10.1055/s-2005-858662

[83] Dietrich CF. Contrast-enhanced low mechanical index endoscopic ultrasound (CELMI-EUS). Endoscopy 2009; 41 (Suppl. 2): E43–44. doi:10.1055/s-0028-1119491

[84] Greis C. Technology overview: SonoVue (Bracco, Milan). Eur Radiol 2004; 14: P11–15

[85] Wei K, Jayaweera AR, Firoozan S et al. Quantification of myocardial blood flow with ultrasound-induced destruction of microbubbles administered as a constant venous infusion. Circulation 1998; 97: 473–483. doi:10.1161/01.cir.97.5.473

[86] Reuter DA, Huang C, Edrich T et al. Cardiac output monitoring using indicator-dilution techniques: basics, limits, and perspectives. Anesthesia and analgesia 2010; 110: 799–811. doi:10.1213/ANE.0b013e3181cc885a

[87] Averkiou MA, Juang EK, Gallagher MK et al. Evaluation of the Reproducibility of Bolus Transit Quantification With Contrast-Enhanced Ultrasound Across Multiple Scanners and Analysis Software Packages-A Quantitative Imaging Biomarker Alliance Study. Investigative radiology 2020; 55: 643–656. doi:10.1097/RLI.0000000000000702

[88] Krix M, Plathow C, Kiessling F et al. Quantification of perfusion of liver tissue and metastases using a multivessel model for replenishment kinetics of ultrasound contrast agents. Ultrasound in medicine & biology 2004; 30: 1355–1363. doi:10.1016/j.ultrasmedbio.2004.08.011

[89] Krix M, Kiessling F, Farhan N et al. A multivessel model describing replenishment kinetics of ultrasound contrast agent for quantification of tissue perfusion. Ultrasound in medicine & biology 2003; 29: 1421–1430. doi:10.1016/s0301-5629(03)01033-0

[90] Krix M, Kiessling F, Vosseler S et al. Comparison of intermittent-bolus contrast imaging with conventional power Doppler sonography: quantification of tumour perfusion in small animals. Ultrasound in medicine & biology 2003; 29: 1093–1103. doi:10.1016/s0301-5629(03)00060-7

[91] Arditi M, Frinking PJ, Zhou X et al. A new formalism for the quantification of tissue perfusion by the destruction-replenishment method in contrast ultrasound imaging. IEEE transactions on ultrasonics, ferroelectrics, and frequency control 2006; 53: 1118–1129. doi:10.1109/tuffc.2006.1642510

[92] Hudson JM, Karshafian R, Burns PN. Quantification of flow using ultrasound and microbubbles: a disruption replenishment model based on physical principles. Ultrasound in medicine & biology 2009; 35: 2007–2020. doi:10.1016/j.ultrasmedbio.2009.06.1102

[93] Dietrich CF, Averkiou M, Nielsen MB et al. How to perform Contrast-Enhanced Ultrasound (CEUS). Ultrasound Int Open 2018; 4: E2–E15. doi:10.1055/s-0043-123931

[94] Wildeboer RR, van Sloun RJG, Huang P et al. 3-D Multi-parametric Contrast-Enhanced Ultrasound for the Prediction of Prostate Cancer. Ultrasound in medicine & biology 2019; 45: 2713–2724. doi:10.1016/j.ultrasmedbio.2019.05.017

[95] Cui XW, Ignee A, Bachmann Nielsen M et al. Contrast enhanced ultrasound of sentinel lymph nodes. Journal of Ultrasonography 2013; 13: 73–81

[96] Cui XW, Ignee A, Jedrejczyk M et al. Dynamic Vascular Pattern (DVP), a quantification tool for contrast enhanced ultrasound. ZGastroenterol 2012; in press

[97] Wiesinger I, Jung F, Jung EM. Contrast-enhanced ultrasound (CEUS) and perfusion imaging using VueBox(R). Clin Hemorheol Microcirc 2021; 78: 29–40. doi:10.3233/CH-201040

[98] Jung EM, Wertheimer T, Putz FJ et al. Contrast enhanced ultrasound (CEUS) with parametric imaging and time intensity curve analysis (TIC) for evaluation of the success of prostate arterial embolization (PAE) in cases of prostate hyperplasia. Clin Hemorheol Microcirc 2020; 76: 143–153. doi:10.3233/CH-209202

[99] Dong Y, Koch JBH, Lowe AL et al. VueBox(R) for quantitative analysis of contrast-enhanced ultrasound in liver tumors1. Clin Hemorheol Microcirc 2022; 80: 473–486. doi:10.3233/CH-211261