

REVIEW

Update on the safety and efficacy of commercial ultrasound contrast agents in cardiac applications

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Abstract

Ultrasound contrast agents (UCAs) are currently used throughout the world in both clinical and research settings. The concept of contrast-enhanced ultrasound imaging originated in the late 1960s, and the first commercially available agents were initially developed in the 1980s. Today's microbubbles are designed for greater utility and are used for both approved and off-label indications. In October 2007, the US Food and Drug Administration (FDA) imposed additional product label warnings that included serious cardiopulmonary reactions, several new disease-state contraindications, and a mandated 30 min post-procedure monitoring period for the agents Optison and Definity. These additional warnings were prompted by reports of cardiopulmonary reactions that were temporally related but were not clearly attributable to these UCAs. Subsequent published reports over the following months established not only the safety but also the improved efficacy of clinical ultrasound applications with UCAs. The FDA consequently updated the product labeling in June 2008 and reduced contraindications, although it continued to monitor select patients. In addition, a post-marketing program was proposed to the sponsors for a series of safety studies to further assess the risk of UCAs. Then in October 2011, the FDA leadership further downgraded the warnings after hearing the results of the post-marketing data, which revealed continued safety and improved efficacy. The present review focuses on the use of UCAs in today's clinical practice, including the approved indications, a variety of off-label uses, and the most recent data, which affirms the safety and efficacy of UCAs.

Key Words

- ▶ ultrasound contrast agents
- ▶ contrast echocardiography
- ▶ contrast-enhanced ultrasound

Introduction

The discovery of contrast-enhanced ultrasound (CEUS) can be dated to 1968, when Gramiak & Shah (1) reported their observations of 'clouds of bubbles' that appeared in the aortic root following injections of saline through an intra-aortic catheter. The pioneering investigators Bove *et al.* (2) and Kremkau *et al.* (3) also noted similar findings, and the concept of CEUS was born. Early investigators, including Meltzer *et al.* (4) and Reale *et al.* (5), subsequently set the stage for the development of ultrasound contrast agents (UCAs) and led to the validation of these agents as true, non-diffusible

intravascular indicators. Once the physical locations of UCAs were defined, investigators focused on utility. DeMaria *et al.* (6) and Ong *et al.* (7) were among the earliest innovators to develop methods for quantifying the acoustic effects of UCAs for cardiac applications, and others, including Armstrong *et al.* (8) and Sakamaki *et al.* (9), associated UCAs with specific clinical conditions. Ultimately, these combined efforts led to the discovery and utility of several different first-generation UCAs.

First-generation UCAs initially included compounds such as agitated saline, Indocyanine green, hydrogen peroxide, and sonicated solutions of dextrose and Renograffin (10). These microbubbles were generally considered



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either too large to pass through the pulmonary circulation or too unstable (10). Therefore, the clinical applications were less than ideal for daily use. Subsequently, Feinstein *et al.* (11) showed that sonicated microbubbles were both small and stable enough to traverse the pulmonary circulation and to opacify the left ventricle (LV). These findings led to significant interest in the development of commercially available UCAs by pharmaceutical companies. The first commercial UCAs became available in the 1980s and included Echovist (1982) and Levovist (1985), which were available in Europe, Japan, and Canada. Alunex, the first commercial agent approved by the US Food and Drug Administration (FDA) was subsequently released in the USA in 1994. Levovist and Alunex microbubbles successfully transited the pulmonary circulation, although the efficacy of these UCAs remained inconsistent because of the fact that the gas component was predominantly nitrogen and, as such, was relatively diffusible (12). The need for a more robust product sparked the production of the agents that are currently used in today's clinical practice, which are often termed second-generation UCAs.

Current clinical UCAs and imaging techniques

Current UCAs consist of microbubbles of an inert, relatively insoluble gas encapsulated by a protein, lipid, or polymer shell. The gas, which is typically perflutren (octafluoropropane) or sulfur hexafluoride, is characterized as a high-density, high-molecular weight gas that exhibits low solubility. The shell of protein or lipid provides enhanced stability, which leads to improved durability and functionality. The microbubbles typically range from 1 to 10 μm in diameter (red blood cell diameter is $\sim 7.8 \mu\text{m}$), which thus permits unhindered passage from the peripheral injection site through the pulmonary vasculature with subsequent entrance into the left heart chambers and access to the systemic circulation. The size of the individual UCAs do not permit passage through the endovascular borders and therefore remain as truly intravascular indicators.

Coupled with external ultrasound imaging systems, the microbubbles induce a dramatic alteration in the acoustic impedance reflection patterns within tissue or blood. The UCAs result in a marked signal-to-noise ratio and provide enhancement of the ultrasound signals, which results in a dramatic improvement in quality. Because of the inherent size of the UCAs and the frequency range of the interrogating acoustic energy, microbubbles exhibit unique vibrational patterns that are represented

by the initial resonance frequency, which includes multiple reflection patterns with behavior that has been described as non-linear. The second frequency, or the second harmonic, is often used to enhance diagnostic images because it has a theoretical advantage over the fundamental frequency, insofar as its adjacent tissue structures do not resonate as microbubbles. Therefore, UCAs markedly improve the signal-to-noise ratio for clinical ultrasound examinations (13). At higher incident pressures (mechanical index > 0.3), microbubble shells are often disrupted. Therefore, it is recommended that a non-linear imaging technique combined with a low harmonic or a very low real-time perfusion (mechanical index < 0.2) pulse sequence scheme be performed for all injections (14). Very low real-time perfusion MI techniques, including pulse inversion Doppler, power modulation, and contrast pulse sequencing, have been developed to improve the visualization of the microbubble response and to eliminate background tissue signals (15).

The first commercialized second-generation UCA produced was Optison (1997). It was subsequently followed by several other competing agents, including Definity (2001), Sonovue (2001), Luminty (2006), Sonazoid (2007), and Lumason (2014). Over the years, multiple other agents were developed, but many are no longer in production. The focus of the present review remains on the commercial contrast agents currently in clinical use (Table 1).

Optison

Optison consists of microspheres of protein-type A microspheres that contain perflutren. It is produced by GE Healthcare (Princeton, NJ, USA) and is currently marketed in both Europe and North America. Optison is typically administered using a bolus via peripheral i.v. injections. As a class, all UCAs remain contraindicated in patients with known right-to-left or bidirectional intracardiac shunts, and Optison is specifically contraindicated in patients with known hypersensitivity to perflutren, blood, blood products, or albumin (16).

Definity and Luminty

Definity and Luminty consist of microspheres with an outer lipid shell that encapsulates perflutren. Produced by Lantheus Medical Imaging (Billerica, MA, USA), they are currently marketed in North America, Europe, Australia, and parts of Asia (17). These agents can be administered by either bolus or infusion. Definity is contraindicated in patients with known right-to-left or bidirectional

Table 1 Currently marketed ultrasound contrast agents.

Brand name	Manufacturer	Year approved	Inner gas	Outer shell	Approved for	Marketed in
Optison	GE Healthcare	1997	Perflutren	Albumin	LVO/EBD	USA, Europe
Definity/Luminity	Lantheus Medical Imaging	2001/2006	Perflutren	Lipid	LVO/EBD, Liver ^a , Kidney ^a , DAV ^a	USA, Canada, Europe, Australia, parts of Asia
Sonovue/Lumason	Bracco Imaging S.p.A	2001/2014	Sulfur hexafluoride	Lipid	LVO/EBD, Breast ^a , Liver ^a , DAV ^a	North America, New Zealand, Europe, Brazil, parts of Asia
Sonazoid	GE Healthcare	2007	Perflubutane	Lipid	Liver ^a , Breast ^a	Japan, South Korea

LVO, left ventricular opacification; EBD, endocardial border definition; DAV, diagnostic assessment of vessels.
^aOnly in certain countries.

intracardiac shunts and those with hypersensitivity to perflutren. Definity has been linked to a very low incidence of non-fatal events that are termed complement activation-related pseudo allergy, and these events may range from temporary back pain to hypotension to hypoxemia to angioedema (18).

Sonovue and Lumason

Sonovue and Lumason consist of microspheres with an outer lipid shell that encapsulates sulfur hexafluoride gas. They are produced by Bracco Imaging S.p.A (Milan, Italy) and are currently marketed in Europe, North America, Australia, and parts of Asia and South America. These agents can be administered by either bolus or infusion. Sonovue is contraindicated for those patients with hypersensitivity to the active substances, patients known to have right-to-left shunts, severe pulmonary hypertension (pulmonary artery pressure >90 mmHg), or uncontrolled systemic hypertension, and patients with acute respiratory distress syndrome. Sonovue should not be used in combination with dobutamine in patients with conditions that suggest cardiovascular instability where dobutamine is contraindicated (19).

Sonazoid

Sonazoid consists of microspheres with an outer lipid shell that encapsulates perfluorobutane gas. It is produced by GE Healthcare and is marketed by Daiichi Pharmaceutical Co., Ltd (Tokyo, Japan). Sonazoid can be administered by either bolus or infusion; however, the usual dosage for an adult is 0.015 ml/kg body weight in a single administration (20). Sonazoid is contraindicated in patients with known egg allergies and has been associated with a low incidence of side effects, such as diarrhea, albuminuria, and neutropenia (21).

Current clinically approved indications and emerging clinical uses

As mentioned earlier in the present report, the aim of the second-generation UCAs was to create a product that was both stable and reliable and that would allow unhindered passage through the pulmonary vasculature, which would thereby permit left ventricular opacification (LVO) and LV endocardial border definition (EBD). The clinical need for such agents has risen, seeing as more than 30% of certain populations use echocardiograms that are considered technically difficult or uninterpretable (22). Several studies have demonstrated the efficacy and safety of these agents in improving the diagnostic utility of both stress and rest transthoracic echocardiography (23, 24, 25, 26). Others have even shown that the administration of UCAs can decrease additional diagnostic testing by up to 33%, can alter pharmacologic management by 10%, and is cost-effective (27).

The current indications for UCAs primarily depend on the approval patterns of individual countries' regulatory agencies. The sole approved indication of UCAs in the USA at this time is for LVO/EBD. The American Society of Echocardiography (ASE) published guidelines for the performance of CEUS by sonographers in 2001 (28), a focused update of these guidelines in 2014 (14), and guidelines for the clinical applications of CEUS in 2008 (29). It recommends that UCAs be used for improved endocardial visualization (i.e., when two contiguous endocardial segments of the LV are not observed or to improve Doppler evaluations if the initial spectral signals are inadequate). It also recommends using UCAs in the following clinical settings (14):

- during situations in which the serial assessment of ejection fraction is required (i.e., chemotherapy) and when visualization of the endocardium is critical

(e.g., the evaluation of chest pain and during stress echocardiography), because UCAs decrease variability and increase accuracy;

- when apical hypertrophic cardiomyopathy and/or LV noncompaction is suspected but not clearly documented or excluded;
- for the assessment of intracavitary thrombi whenever the LV apex is not clearly visualized on a patient with severely depressed systolic function; and
- to help further define LV aneurysms and pseudoaneurysms and to characterize Takotsubo cardiomyopathy.

In Europe, UCAs have other approved indications beyond LVO/EBD, including the detection and characterization of liver and breast masses as well as for Doppler enhancement and the assessment of vasculature. Furthermore, UCAs are often administered for several off-label uses. These off-label uses are frequent and are so well studied that the European Federation of Societies for Ultrasound in Medicine and Biology (EFSUMB) published guidelines in 2012 on the practice of using CEUS in non-hepatic applications (30). In these guidelines, the EFSUMB gives recommendations for using off-label CEUS for pancreatic masses, contrast-enhanced endoscopic ultrasounds, the evaluation of the GI tract and spleen, the evaluation of the entire urogenital tract, and much more. In Canada, Definity is approved for LVO/EBD and also for imaging of the liver and kidney. In Japan, Sonazoid is currently indicated for the evaluation of focal liver and breast lesions.

Today's use of UCAs in diagnostic imaging continues to extend beyond the currently approved indications and those that were published by the EFUSMB in 2012. Presently, UCAs are being used in the evaluation of endovascular repair to detect endoleaks as well as in carotid artery imaging to improve both the detection of neovascularization within carotid plaque and the identification of vulnerable plaque. UCAs are frequently used during transesophageal echocardiograms (TEEs) to further define left atrial appendage (LAA) anatomy and to 'rule out' thrombus before electrophysiologic procedures or to evaluate for strokes. UCAs are also used to better define the cardiac chambers for the three-dimensional analysis of LV function, and they have shown the potential for use in myocardial deformation analysis, although this latter use is currently controversial and not well supported.

Beyond these applications, UCAs exhibit the potential for therapeutic applications, including targeted thrombolysis and drug/gene delivery. Clinical trials are currently underway to test whether UCAs and alterations in the

mechanical index setting can improve outcomes in acute ST segment elevation myocardial infarction by inducing thrombus dissolution as well as work in animals on intracranial thrombi. UCAs are also being used extensively in research studies to help facilitate drug and gene delivery by a process known as sonoporation. Drugs and genes may be loaded or bound to a microbubble or administered separately following i.v. drug infusion, and upon insonation, they can be released and driven into the targeted tissue. Kotopoulos *et al.* (31) have shown in a small group of patients with pancreatic cancer that sonoporation induced a reduction in tumor size and a decrease in mortality. Although the possibility of using UCAs in drug and gene delivery is an exciting new frontier, it is also vast and beyond the scope of the present article.

Safety concerns and the efficacy of UCAs

In October 2007, the FDA issued a new three-part product labeling revision for both Optison and Definity. These labeling changes were issued largely based on reports of four patient deaths and ~190 other serious cardiopulmonary reactions that were temporally related but not clearly caused by UCAs (32). The revision included a 'black box warning', which indicated that serious cardiopulmonary reactions, including fatalities, had occurred during or within 30 min following the administration of UCAs, multiple new disease-state contraindications to UCAs, and a 30 min monitoring period that included the measurement of vital signs and ECG in all patients and pulse oximeter monitoring for patients at risk for hypoxemia. These revisions had an astounding negative impact on UCA use in the USA, with marketing data for the 2008 calendar year showing that UCAs were only used in 3.2% of stress echocardiograms and in 0.4% of resting echocardiograms; both percentages were markedly lower than those from 2007 (33). Many physicians, including those who organized the International Contrast Ultrasound Society (ICUS), were openly critical of the FDA's official actions; they cited the prodigious amount of existing safety and efficacy data regarding the use UCAs and the lack of a proven causal relationship between the reported adverse events and UCA administration (34, 35).

In early 2008, in response to the FDA revisions, several investigators began to publish on the safety and improved efficacy of UCAs. Main *et al.* (36) showed that in a retrospective, propensity-matched population, consecutive patients that underwent an echocardiogram were 24% less likely to die within 1 day than were patients who did not receive a contrast agent. Wei *et al.* (37) also noted

an incredibly low serious adverse event (SAE) rate of 0.01% in those patients that received UCAs. Several others also noted the safety of these agents in stress echocardiograms as well as the lack of increased events in long-term follow-up.

By May 2008, following the publication of several trials focused on safety data that were accompanied with passionate lobbying by physicians, the FDA announced significant product label revisions for both Optison and Definity. Although the anemic black box warning remained, disease-state contraindications were modified to warnings, and the mandated 30 min monitoring period was lifted for all patients except those with pulmonary hypertension and unstable cardiopulmonary conditions. In June 2008, the FDA, in conjunction with the manufacturers of both Optison and Definity, released details on a post-marketing program that included three separate safety studies to further assess the risk of each UCA. These studies included a retrospective observational study that used a large administrative database to determine the risk of mortality in critically ill patients who undergo echocardiography with or without an UCA, a prospective invasive pulmonary hemodynamic study in patients with normal or elevated baseline pulmonary artery systolic pressure, and a prospective multicenter safety registry in ~1000 patients (38). The results of these trials showed that there were no significant increases in mortality, no changes in pulmonary artery pressures, and no SAEs (Table 2).

In October 2011, after reviewing the preliminary data from the risk mitigation studies, the FDA officials made the most recent modification to the package insert for Definity; in it, they removed the monitoring requirements for patients with pulmonary hypertension or unstable cardiopulmonary conditions, and they added the notation that severe reactions to the agents occur uncommonly.

Also, the statement that the efficacy and safety of these agents had not been established in stress testing was removed. They subsequently made a similar change to the package labeling of Optison in March 2012. The proposed risk mitigation studies have all subsequently been published in peer-reviewed journals.

Sonovue has been commercialized across Europe and Asia since 2001. Upon its initial release, the Committee of Human Medicinal Products (CHMP), a division of the European Medicines Agency, labeled the product as contraindicated in known right-to-left shunts, unstable coronary syndromes, congestive heart failure, severe pulmonary hypertension, and pregnant or breastfeeding patients. In June 2014, following an extensive evaluation by the CHMP of the benefits and risks of Sonovue in critically ill patients, the decision was made to remove the contraindication for use in patients with recent acute coronary syndrome or clinically unstable ischemic heart disease. CHMP added that Sonovue should not be used in combination with dobutamine in patients with conditions that suggest cardiovascular instability where dobutamine is contraindicated (39).

Update on UCA safety and efficacy data: March 2012 to present

Since the time of the most recent change to the package labels for both Optison and Definity, there have been several published articles and reviews detailing both the safety and efficacy of UCAs in a variety of patient populations. In 2012, Wever-Pinzon *et al.* (40) published a retrospective series on 1513 consecutive inpatients with documented pulmonary hypertension who had received UCAs. These patients were followed for 24 h post-UCA administration for SAEs, including respiratory decompensation, hypotension, arrhythmias, syncope,

Table 2 FDA mandated post-marketing safety studies. Data available at <http://www.fda.gov/downloads/AdvisoryCommittees/CommitteesMeetingMaterials/Drugs/CardiovascularandRenalDrugsAdvisoryCommittee/UCM256586.pdf>^a.

	Retrospective database of critically ill patients	Pulmonary hemodynamics	Prospective safety registry
Definity	n = 15 775 propensity matched patients HR = 0.71 (0.62–0.82) (47)	n = 32 No change in mean PAP (48)	n = 1053 No deaths or SAE at 24 h (49)
Optison	n = 2900 propensity matched patients HR = 1.18 (0.82–1.71) (50)	n = 30 No change in mean PAP (51)	n = 1039 No deaths or SAE at 24 h (52)

PAP, pulmonary artery pressure; SAE, serious adverse events.

^aAdapted from Boolani H & Main M. Update on contrast echocardiography: safety and utility. *Current Cardiovascular Imaging Reports* 2012 5 410–419.

convulsions, anaphylactic reactions, or death. Out of the 1513 patients, only three patients had an SAE after the administration of the UCA, and none of these events was directly attributed to the UCA itself. Goldberg *et al.* (41) conducted a retrospective, single-center study that involved 96 705 transthoracic echocardiograms, and Definity was used in 2518 of them. They found that overall mortality at 24 h was 0.44% in the Definity group and 0.69% in the non-contrast group ($P=0.14$). Multivariate analysis showed that the administration of Definity was not associated with increased mortality after adjustment for age, sex, race, patient location, ejection fraction, and the presence of various comorbidities ($P=0.67$).

In 2013, Platts *et al.* (42) performed a multicenter retrospective analysis on consecutive patients in Australia who received Definity and were monitored for SAE over a 5-year period. A total of 5956 studies with UCAs were performed, most of which were outpatient stress tests. There were 16 SAEs related to UCAs (0.27%), all of which were mild and transient, with the most common reactions being back pain and rash. There were no cases of serious anaphylaxis or death within 30 min of the contrast administration. Subsequently, ICUS released a review on the safety of UCAs in patients with known cardiac shunts that focused on the physiology of UCAs and compared them to other commonly used i.v. radiopharmaceuticals. They recommended the removal of the contraindications in order to further the public interest in safe, reliable, radiation-free diagnostic imaging options for patients with known or suspected cardiac shunts and to reduce the need for unnecessary downstream testing of the UCAs.

Also in 2013, several authors investigated the application and ability of perfusion myocardial contrast echocardiography (MCE) to detect coronary artery disease (CAD) and to predict outcomes. Anantharam *et al.* (43) published a letter to the editor showing that perfusion contrast echocardiography, when it was used to diagnose ischemic burden in patients that initially present with heart failure, was an independent predictor of mortality, with an area under the ROC curve of 0.67. Although their sample size was small ($n=89$), that study was the first to show that the extent and severity of ischemia as determined by contrast echocardiography is predictive of all-cause mortality. Porter *et al.* (44) subsequently performed a prospective single-center randomized controlled trial to evaluate the effectiveness of MCE vs conventional stress echo. They showed that an abnormal MCE was more frequently observed than an abnormal conventional stress echo was ($P<0.001$), and it more frequently resulted in

revascularization ($P<0.004$). Senior *et al.* (45) performed a multicenter prospective study on patients undergoing MCE, single-photon emitted computed tomography (SPECT), and coronary angiogram. They found that UCA MCE demonstrated superior sensitivity but lower specificity for the detection of CAD as compared to SPECT in a population with a high incidence of CV risk factors and an intermediate to high prevalence of CAD.

In 2014, Klara *et al.* (46) reported on the safety of UCAs in right-to-left intracardiac shunts in a letter to the editor that followed a publication by the ICUS on the safety of known or suspected cardiac shunts. They showed that in a retrospective analysis of 418 consecutive patients with a known right-to-left intracardiac shunt who had received UCAs, there were no primary adverse events (including neurologic or embolic phenomena) and only one secondary adverse event (back pain). Patients with known left-to-right shunts ($n=63$) were excluded. These numbers were not statistically different from all of the other patients who received UCAs, and the study's authors suggested that the contraindication of UCAs in patients with known intracardiac shunts be rescinded. The updated August 2014 focused guidelines for contrast use by the ASE clearly denounce the purported increased risk of i.v. commercial contrast agents in patients known to have small right-to-left shunts through a patent foramen ovale (PFO) (saline contrast in the left atrium (LA) or LV that is transient and does not fill the LA or LV cavity) (14). Also in 2014, Shah *et al.* (54) showed that MCE provided incremental benefit over wall motion analysis in 25% of patients undergoing stress echocardiography and greater confidence with wall motion analysis in 62%. MCE detected significantly more cases of ischemia and detected a greater ischemic burden than did wall motion analysis on a per patient basis.

In conclusion, UCAs have time and time again shown their ability not only to reduce intra- and interobserver variability in echocardiography interpretation but also to reduce upstream testing, medical costs, mortality, and exposure to the ionizing radiation that is associated with other imaging modalities. And yet although their applications in research and off-label indications are growing almost exponentially, the UCAs remain significantly underutilized in today's routine clinical echocardiography practice. As increasing support mounts for UCA use with publications about the safety and efficacy of UCAs along with the benefits of using UCAs in many other non-cardiac applications, we remain hopeful that the advantages will become widely known and accepted by clinicians and researchers alike. We remain optimistic

that the manufacturers of UCAs will continue to pursue additional clinical applications in order to provide safe, efficacious, and valuable diagnostic and therapeutic options for improving the health and well-being of our patients.

Declaration of interest

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