SCIENTIFIC DISCUSSION

1. Introduction

Echocardiography

Over the past decade, echocardiography has become an imaging method of choice for evaluating individuals with suspected cardiac disorders, largely because of its ease of use, widespread availability, and portability.
Despite the many positive features of echocardiography, poor diagnostic quality images are still obtained in many patients. It is estimated that as many as 10 to 15% of echocardiograms using fundamental imaging do not provide diagnostically acceptable visualisation of the left-ventricular (LV) endocardial borders. Although new ultrasound technologies such as harmonic imaging have dramatically improved endocardial border delineation, it remains inadequate in a substantial number of patients. Echocardiographic assessments of regional and global ventricular function are consequently limited or impossible in many patients. The inability to clearly identify the ventricular cavity and endocardial borders can result in a failure to detect regional and global left ventricular function, in poor reader's confidence and in non-diagnostic examination. This may be overcome by the use of contrast-enhancing agents.

About the product

The principle of the first contrast-enhancing agents was to produce microscopic gas bubbles by shaking, for example, air in a saline, which, when injected gave rise to altered echoes from blood or tissue. These appeared to be too unstable or too large to pass through the pulmonary circulation and led to the development of stabilised and miniaturised air mirocrospheres.
Luminity (DMP115) consists of phospholipid-encapsulated perflutren (perfluropropane, PFP, octafluoropropane) microspheres. It belongs to a third generation of contrast-enhancing agents composed a high-molecular-weight gases, less diffusible than air, stabilised with an outer layer of protein, lipids or synthetic polymers. Similarly to the second generation microspheres, they can pass through the pulmonary circulation and reach the cardiac chambers. Moreover, gases of high molecular weight are not very soluble, which increases in vivo stability of the microspheres and enhances blood echogenicity.

There are two contrast enhancing agent with an echocardiography indication authorised through the centralised procedure and currently available in Europe: SonoVue (EU/1/01/177/01-02) and Optison (EU/1/01/166/01-02).

The present application for marketing authorisation of Luminity is made under Article 8.3 of Directive 2001/83/EC, as amended.

The approved indication is "This medicinal product is for diagnostic use only. Luminity is an ultrasound contrast-enhancing agent for use in patients in whom non-contrast echocardiography was suboptimal (suboptimal is considered to indicate that at least two of six segments in the 4- or 2-chamber view of the ventricular border were not evaluable) and who have suspected or established coronary artery disease, to provide opacification of cardiac chambers and improvement of left ventricular endocardial border delineation at both rest and stress

The applicant also sought approval "for contrast-enhanced ultrasound imaging in patients referred for diagnostic ultrasound imaging to improve the characterization of focal lesions of the liver and kidney". During the scientific assessment procedure, the applicant withdrew the claim for this indication. At the time of withdrawal, the CHMP considered that the clinical data provided were not sufficient to establish the clinical efficacy of Luminity in this indication, since there were unresolved major objections on the demonstration of clinical efficacy.

2. Quality aspects

Introduction

Before activation, Luminity is presented as a vial containing a colourless, uniformly clear to translucent liquid with perflutren gas in the headspace.
After activation, Luminity is presented as a dispersion for injection or infusion with perflutren-containing lipid microspheres as active substance. Since the diagnostic activity is dependent upon the number and the size of the dispersed gas microspheres, it is relevant to state that each millilitre of dispersion contains a maximum of $6.4x10^9$ microspheres, with a mean diameter range of 1.1-2.5 micrometres. The approximate amount of perflutren gas in each millilitre of Luminity is 150 microlitres.

The other ingredients include 1,2-dipalmitoyl-*sn*-glycero-3-phosphatidylcholine (DPPC), 1,2-dipalmitoyl-*sn*-glycero-3-phosphatidic acid, monosodium salt (DPPA), N-(methoxypolyethylene glycol 5000 carbamoyl)-1,2-dipalmitoyl-*sn*-glycero-3-phosphatidylethanolamine, monosodium salt (MPEG5000 DPPE), sodium dihydrogen phosphate monohydrate, disodium hydrogen phosphate heptahydrate, sodium chloride
propylene glycol, glycerol and water for injections.

Luminity is packed in clear borosilicate Type I glass vials, closed with a chlorobutyl elastomeric stopper, and sealed with an aluminium crimp seal having a plastic flip-off button. Before use Luminity has to be activated using a mechanical shaking device (Vialmix), not part of the presentation of the medicinal product but to be provided separately to healthcare professionals by Bristol-Myers Squibb Pharma Belgium Sprl.

Active Substance

For simplicity, the active substance may be said to be perflutren. However, it may be more correct to refer to the dispersed system of perflutren-containing lipid microspheres as the echogenic contrast enhancement is related to a number of physical parameters of the system as a whole – number of microspheres/ml, size, properties of perflutren/lipids interface etc…

Perflutren has the molecular formula C_3F_8. It is a non-polar, colourless and odourless gas. It is chemically inert and thermally stable (decomposition > 400°C) because of the strength of the carbon-fluorine bond. Detailed information on quality/control of materials used in the synthesis, as well as on the synthesis itself, has been provided by the way of an active substance master file.

- Manufacture

Perflutren is synthesised via a 3-stage process consisting of one synthetic and two purification steps. Satisfactory specifications and associated methods have been provided for the starting materials, intermediate, reagents and solvents. Validation data submitted confirm robustness and reproducibility of the process.
Neither class 1 or class 2 solvents nor metal catalysts are used in the process. A discussion about potential impurities including their origin according to the manufacturing process is provided. Their content appear to be very low.

- Specification

The active substance specification include test for identity (IRFT), assay (GC), purity, related substances (GC), air, residual free fluoride and heavy metals. Impurity limits in the specification are justified by toxicology studies.

Batch analysis data provided for 21 batches of perflutren manufactured at the commercial site confirm satisfactory compliance and uniformity with the proposed specification.

- Stability

Stability data have been provided for four batches. Three batches were first subjected to six months storage under accelerated conditions (40°C/75% RH) and then moved to the long term study. The fourth batch was directly included in the long-term stability study (15-30ºC and ambient humidity to 80%). 6-month data and up to 3-year data are available respectively under long term and accelerated conditions.
The parameters tested included assay, impurities, air, container weight, residual fluoride and heavy metals.
The stability studies were not conducted fully in line with EU Guidelines. However, in view of the chemical inertness of perflutren and the impermeable nature of the gas cylinders to be used as containers, it was considered in this special case that the data presented support the proposed retest period when perflutren is stored in the proposed packaging

Medicinal Product

- Pharmaceutical Development

The performance of an ultrasound contrast-enhancing agent is determined by its physical characteristics i.e. size (range of greatest interest 1 to 10 micrometre), number, and stability of the microspheres in the finished product. The aim of the development was to overcome the inherent instability and consequential short duration of action of the first generation of ultrasound contrast agents (see introduction).

Perflutren has been selected as the dispersed gas in the microspheres because of its extremely low water solubility increasing the stability of the microspheres, chemical inertness minimising the risk of interactions, and availability as a highly purified material.
Phospholipids were chosen to stabilise the perflutren microspheres because they are: endogenous to biological systems (DPPC and DPPA are major constituents of human cell membranes), commercially available from non-animal sources and amphiphilic (formation of microspheres in aqueous systems readily achievable). MPEG5000 DPPE, for which satisfactory data on composition, manufacture, control and toxicology have been provided (see non clinical section), has been selected to minimise recognition by the reticuloendothelial system. DPPA, imparting a charge to the microspheres, was chosen to reduce coalescence. The ratio of the three lipids has been optimised during development with regards to microsphere size distribution and presence of unincorporated phospholipids. The phosphate buffer has been incorporated in the formulation for phospholipids stability purposes. Sodium chloride is used as a tonicity agent, propylene glycol to improve the wetability of the phospholipids during compounding, and glycerol to increase the viscosity, which is believed to enhance the microspheres stability.
The microspheres have been satisfactorily characterised mainly in term of size distribution before and after activation, structure, lipid envelopment of perflutren and coalescence.
All the excipients are controlled according to satisfactory specification. The finished product does not contain any component of animal/human origin. The phospholipids are incorporated during the manufacture of the finished product as a lipid blend, for which satisfactory data have been provided concerning description/validation of the manufacturing process, specification, batch analysis data, description and validation of analytical methods and stability data/retest period.

Satisfactory specifications have been provided for the type I borosilicate glass vials treated with ammonium sulphate and the chlorobutyl elastomeric stoppers. This type of vial was selected as primary packaging material as exposure of untreated glass surfaces to phosphates is known to delaminate silica and to generate particles. The integrity of the seal, especially with regards to the perflutren gas content in the headspace, has been confirmed by a dye ingress test and by stability data.

Satisfactory *in vitro* and *in vivo* data have been provided in support of chemical and acoustic equivalence of the 3 different formulations used in clinical studies (see clinical section).

When administered by infusion Luminity may be diluted with sodium chloride 9 mg/ml (0.9%) solution for injection or glucose 50 mg/ml (5%) solution for injection. This results in a fall in concentration perflutren microspheres, which can be overcome by increasing the infusion flow rate, thereby maintaining the image quality (see SPC section 4.2). No significant effect on microsphere distribution was observed when samples were exposed to flow rates and needle diameters (18 and 20 gauge needles) of clinical relevance indicating the microspheres are deformable.

Before use, Luminity has to be activated with a mechanical shaking device (Vialmix) not part of the presentation of the medicinal product but to be provided to healthcare professionals by Bristol-Myers Squibb Pharma Belgium Sprl. The duration and frequency of oscillation of the shaker has been set up so that acceptable fraction of microspheres falls within the clinically important range. It was shown that the concentration of large microspheres will not increase unacceptably if the activation period is prolonged or if the oscillation frequency deviates from the target. If directly administered to patients without activation, the product will not produce its intended effect, because only the gas dispersion is echogenic. The vialmix is designed to cut off after the set up optimum activation time (45 seconds) and in case the activation process is interrupted before completion, an audible alarm and a visual display message will activate.

- Manufacture of the Product

The method of manufacture involves the following operations: dissolution of the lipid blend, addition of the remaining excipients, dilution, sterile filtration, filling, stoppering, transfer of the vials in a lyophiliser where the vacuum created is released with sterile perflutren gas, sealing of the previously stoppered vials with an aluminium seal, terminal sterilisation by autoclaving and packaging.
The conditions of the terminal sterilisation have been slightly modified compared to the PhEur reference conditions in order to minimise potential hydrolytic degradation of the phospholipids. Nevertheless, the chosen sterilisation cycle has been validated and it has shown a satisfactory lethality. Satisfactory in-process controls have been defined.
Validation data provided for three consecutive full-scale batches confirm robustness and reproducibility of the manufacturing process.

- Product Specification

Before activation, the product specification includes tests controlled by validated methods for appearance, container/closure, particulates, perflutren identity (IR), perflutren assay (GC), DPPC assay (HPLC), DPPA assay (HPLC), MPEG5000 DPPE assay (HPLC), degradation products (RP HPLC), content uniformity (PhEur), volume, pH, sterility and endotoxins.
After activation, the product specification includes tests controlled by validated methods for extractable volume (PhEur), stereo microscopy, microsphere size distribution, microspheres concentration, perfutren volume (GC).
The microsphere size distribution specification has been satisfactorily justified in terms of safety and efficacy and it reflects batches used in non-clinical and clinical studies. Stereo microscopy is used to ensure that the number of large microspheres is limited.
Batch analyses data provided for three batches manufactured at the commercial manufacturing site confirm compliance with the specifications and indicate consistent and reproducible manufacture.

- Stability of the Product

Stability of the Product before activation

Stability data have been provided for 3 batches manufactured at the commercial manufacturing site. Under long-term conditions (5°C±3°C - commercial packaging – upright and optional inverted position) and under 2-year data have been provided. Under accelerated conditions (25°C/40% RH - commercial packaging - upright and optional inverted position) 6-month data have been provided.

The parameters tested included appearance, particulates, perflutren identity (IR), perflutren assay, perfutren volume, DPPC assay (HPLC), DPPA assay (HPLC), MPEG5000 DPPE assay (HPLC), degradation products, stereo microscopy, microspheres size distribution, pH, sterility and endotoxins.
The observed changes were small, and not likely to have a significant effect on efficacy and safety of the product when used according to the directions in the SPC
Photostability studies have shown that the finished product is non-light sensitive.
The data provided support the proposed shelf life and storage conditions as defined in the SPC.

Stability of the Product after activation

It has been shown that the microspheres remain 'active' for 12 hours after activation, sufficient to allow image enhancement in vivo.
Studies investigated the effect of reactivation after initial activation on the microspheres characteristics and the results show that a second activation is possible within 48 hours and that the product may be used up to 12 hours after the second activation.
The data provided support the proposed shelf life and storage conditions as defined in the SPC.

Discussion on chemical, pharmaceutical and biological aspects

The active substance is well characterised, documented and it is very stable The pharmaceutical development has focused on selecting a product giving a uniform and reproducible microsphere size distribution characteristics following activation. Some of the excipients are compendial and satisfactory data have been provided in support of the non-compendial ones. The packaging material is well documented. The manufacturing process enhances to obtain reproducible finished product batches. Stability tests under ICH conditions indicate that the product is stable for the proposed shelf life. Moreover, a satisfactory post-activation in-use shelf-life has been established. At the time of the CHMP opinion, there were a number of minor unresolved quality issues having no impact on the benefit-risk balance of the product. The applicant committed to resolve these as Follow Up Measures after the opinion, within an agreed timeframe.

3. Non-clinical aspects

Introduction

The applicant has developed *in-vitro* and used *in-vivo* models in mouse, rats and dogs to study the pharmacodynamic effects of Luminity as a contrast agent used in conjunction with ultrasound in order to aid in the diagnosis and assessment of myocardial perfusion defects. Pivotal studies were conducted according to GLP guidelines.

Pharmacology

Luminity is a lipid-encapsulated perflutren filled microsphere dispersion. It contains a blend of 3 lipids dipalmitoylphosphatidic acid (DPPA), dipalmitoylphosphatidylcholine (DPPC) and dipalmitoylphosphatidylethanolamine polyethylene glycol (MPEG5000-DPPE).

- Primary pharmacodynamics

An *in vitro* testing system was developed that allowed the acoustic attenuation of activated Luminity to be measured at ultrasonic energy, frequencies and pulse repetition rates similar to those used clinically. Ultrasound video intensity was directly related to the concentration of activated Luminity. *In vitro* activated Luminity was significantly more stable to ultrasound exposure when diluted in blood than in saline. Further, the rate of disappearance of contrast agent in blood or saline was related to the acoustic output of the ultrasound scan head and to increases in the ambient pressure. A comparison of microspheres made with the lipid composition of Luminity and either PFP gas or air indicated that both had strong ultrasound contrast.

Studies were performed in the anaesthetized open-chest dog to assess various ultrasound contrast modes as well as to compare bolus and infusion of different dose levels of activated Luminity. The results showed that activated Luminity allowed effective assessment of left ventricular opacification (LVO) with fundamental imaging when given at an optimal level as an intravenous bolus (3-10μL/kg) or 2 minute (10 μl/kg) administration. Similarly myocardial perfusion imaging was achieved using gated second harmonic imaging with activated Luminity administration at the same optimal dose levels. Data with intravenous (IV) administration at a dose of 9 μl/kg/min in saline demonstrated steady state, homogeneous enhancement without ventricular shadowing over 30 minutes. These findings were extended by examination of myocardial perfusion patterns in dogs following coronary occlusion and reperfusion. Accurate detection of induced myocardial perfusion defects was achieved with activated Luminity administered over 2 minutes at 10 μl/kg using gated second harmonic imaging. Similarly, activated Luminity administered over 2 minutes at the optimal dose of 3 μL/kg allowed both gated second harmonic pulse imaging and gated harmonic angio pulse imaging to accurately detect these perfusion defects.

The use of activated Luminity for assessment of microcirculatory perfusion has been indicated by a number of collaborative studies and published reports. The ability of activated Luminity to improve accuracy of Doppler ultrasonographic diagnosis of testicular ischemia was demonstrated in a canine model where the testicular artery was ligated. An intravenous injection of 10-25 μL/kg was shown to improve duplex sonographic evaluation [1]. Activated Luminity has also been shown to have potential in monitoring placental flow in gravid baboons and myocardial opacification in rhesus monkeys. Activated Luminity may also be useful for early detection of metastatic focal lesions in the liver [2] . In this study, the contrast enhancement of the VX-2 tumors correlated well with the angiographic and histopathological appearance of the tumor and the borderline of the tumour could be clearly delineated from the surrounding liver parenchyma.

- Secondary pharmacodynamics

No secondary pharmacodynamic studies have been performed.

- Safety pharmacology programme

Safety pharmacology studies in the dog demonstrated that doses up to 25 times the clinical dose (0.5 ml/kg) did not affect cardiopulmonary function. At higher doses marked increase in respiration rate and pulmonary arterial pressure were observed. However, even in a model where severe pulmonary compromise was induced, the highest dose tested (0.2 ml/kg [10 times the clinical dose]) did not affect cardiopulmonary function. Testing of Luminity 12 hr after activation, containing a lower PFP content indicated even lower cardiopulmonary toxicity. In addition to the total dose administered, a study in rats suggested the rate of activated Luminity administration also influenced the occurrence of clinical signs. Examination of the affect of activated Luminity on the microcirculation indicated a very small fraction (1.2%) of the administered dose was retained and this transient retention did not have a detrimental effect on the systemic hemodynamics even at 40 times the clinical dose (800 μl/kg). Furthermore the lack of any histopathological signs of brain damage following carotid artery administration of activated Luminity to rats at five times the clinical dose (100 μl/kg) supports the lack of effect on the microcirculation. Additional safety of activated Luminity was demonstrated in the rhesus monkey where cardiovascular and ECG effects were not seen even at 50 times the clinical dose (1000 μl/kg). ImaRx collaborative studies with poorly characterized Luminity indicated administration up to 1 ml/kg did not affect the central nervous system (CNS), respiratory system or cardiovascular system. Further, activated Luminity did not affect urinary output or electrolyte balance. *In vitro* studies with blood cells also showed activated Luminity had no effect on histamine release, production of reactive oxygen species or platelet aggregation in rabbits and humans.

- Pharmacodynamic drug interactions

No pharmacodynamic drug interaction studies have been performed.

Pharmacokinetics

Pharmacokinetic studies were aimed at evaluating the distribution and clearance of the components of activated Luminity microspheres. Two of the three phospholipids are endogenous (DPPC and DPPA) and the amount administered in activated Luminity represents a minor amount. Pharmacokinetic studies were therefore based on the non-endogenous components. The MPEG5000 DPPE component of Luminity is not an endogenous phospholipid (although DPPE is) and therefore, its distribution and metabolic fate was followed using ^{14}C-MPEG5000 DPPE in conscious Sprague Dawley rats. Similarly, the gas component of the PFP filled microspheres was followed using a validated gas chromatographic method. Blood and expired air levels of PFP were measured following intravenous administration of activated Luminity in dogs.

- Pharmacokinetics of MPEG5000 DPPE

The pharmacokinetics, tissue distribution and elimination of total radioactivity following an intravenous bolus injection of ^{14} C-MPEG5000 DPPE incorporated in activated Luminity were determined in Sprague-Dawley rats following a 1 ml/kg IV injection (0.9 µCi/kg). The rat was chosen for these studies because of its use in toxicity studies and the flexibility to allow tissue distribution to be determined in replicate at multiple time points. ^{14}C-MPEG5000 DPPE exhibited a bi-phasic clearance from the blood with a terminal half-life of 10.6 hr and a small volume of distribution. By 72 hr, the majority of radioactivity (71.5%) had been cleared in the urine with no significant retention in any of the tissues. At 4 hr post dose, the highest concentrations of activity were in the liver (17.8 % of the injected dose) and plasma (17.2% of the injected dose). At 1 hr post-injection, 80% of the dose was present in the plasma with 60% of the activity in the form of ^{14} C-MPEG5000 lysophosphatidyl ethanolamine (LPE) (produced by hydrolysis of a single palmitic acid). The metabolite profile in the urine at 4 hr showed 90% of the radioactivity in the form of ^{14} C-MPEG5000, indicating loss of the phospholipid. Radioactivity in the feces was below the limit of detection for metabolic profiling.

- Pharmacokinetics of perflutren

The *in vivo* kinetics of the PFP component of activated Luminity was studied following single intravenous administration in male beagle dogs. Following administration of activated Luminity, PFP concentrations in blood and expired air were measured. Limited sensitivity of the gas chromatographic method precluded determination of the blood PFP levels following administration of low and mid doses of activated Luminity. At the high dose level of 1000 µl/kg, peak blood concentrations were in the range 0.0258 to 0.0452 µL PFP/ml. T_{max} ranged from 10 to 45 seconds post-dose. The mean AUC was estimated to be 3,237 nl•sec/ml with a mean elimination half-life of 61 seconds and mean systemic clearance of ~ 19 ml/(kg•sec). The volume of distribution was estimated to be 1,720 ml/kg. A linear one-compartment analysis appeared to best fit the data. The large volume of distribution is attributable to the rapid PFP clearance from the blood without attaining a steady state equilibrium in the blood.

At the middle and high doses, an initial rapid elimination of PFP was observed in the expired air with plateau levels attained by 2 or 4 minutes, respectively. Total PFP eliminated in expired air over the course of the study for these groups was determined to be ~ 117% of the theoretical total dose administered indicating that all of the material was excreted from the body. Mean lung clearance at the high dose was determined to be 24.2 ml/(kg•sec).

Toxicology

Toxicology studies included acute (single dose) and subchronic (up to 1 month in duration) studies in rats, dogs and non-human primates, a battery of *in vitro* and *in vivo* genetic toxicology assays, and developmental and reproductive studies in rats and rabbits. Other toxicology studies to evaluate the potential irritancy and antigenicity of activated Luminity were also performed. Additional toxicology studies in rats and non-human primates were performed to investigate clinical symptomatology observed following high doses of activated Luminity.

- Single dose toxicity

The clinical signs observed in rats, dogs, and cynomolgus monkeys given single intravenous doses of activated Luminity were decreased activity, increased respiration and change in heart rate. They occurred during or soon after intravenous dosing and, apart from mortality, typically resolved within 30 minutes after dosing. In studies conducted by the Applicant, with well-characterised Luminity, the no observable-effect dose (NOEL) for clinical signs was 0.1 ml/kg in rats and 1 ml/kg in cynomolgus monkeys given a single intravenous dose of activated Luminity. The NOEL for clinical signs in dogs given a single intravenous dose of activated was 0.1 ml/kg. No deaths occurred in dogs given doses ≤ 5 ml/kg in these studies.

The no-effect doses for clinical signs in rats, dogs and monkeys are 5, 5, and 50 times, respectively, the recommended maximum clinical dose of 0.02 ml/kg for ultrasound imaging (0.01 ml/kg with possible administration of a second dose of 0.01 ml/kg). When these no-effect doses are expressed in terms of the rate of dose administered, (~ 2 ml/kg/min in rats, ~ 2 ml/kg/min in dogs and ~ 1.2 ml/kg/min in monkeys), the margin is ≥ 60-fold compared to the injection rate for diagnostic use in humans.

- Repeat dose toxicity (with toxicokinetics)

In general, the spectrum of clinical signs observed in rats, dogs, and cynomolgus monkeys given single or repeated daily intravenous injections of activated Luminity were similar. An exception is that only dogs exhibited erythema. As with single doses, the clinical signs with repeat dosing occurred during or soon after intravenous administration and, apart from mortality, typically resolved within 30 minutes in all species. In definitive 1-month toxicity studies conducted by the Sponsor using well-characterised product, the no-observable-effect dose of activated Luminity for clinical signs was 0.1 ml/kg/day in rats and 0.3 ml/kg/day in cynomolgus monkeys. Transient clinical signs were inconsistently observed (not always on consecutive days) in individual dogs given doses of 0.01 ml/kg/day in a 1-week toxicity study. Symptoms were also seen in dogs receiving non-activated Luminity. A no-observable-effect dose was not defined in dogs. The no-observable-effect level (NOEL) for clinical signs after 1 month of treatment in rats (0.1 ml/kg/day) and monkeys (0.3 ml/kg/day) are 5 and 15 times, respectively, the recommended maximum clinical dose of 0.02 ml/kg for ultrasound imaging (0.01 ml/kg with possible administration of a second dose of 0.01 ml/kg). When these no-observable-effect levels are expressed in terms of the rate of dose administered, (2 ml/kg/min in rats and 1.2 ml/kg/min in monkeys), the margin is ≥ 60-fold compared to the injection rate for diagnostic use in humans (0.01 ml/kg administered in 30-60 seconds or 0.01-0.02 ml/kg/min). NOEL for clinical signs for each species and the safety margin relative to the clinical dose is given in the following table.

While lung lesions were observed in rats given ≥ 0.1 ml/kg/day of activated Luminity for 1 month, no lung lesions were observed following a single intravenous dose ≤ 0.3 ml/kg in rats or at the tolerated doses (≤ 1 ml/kg) in cynomolgus monkeys (doses which are 15 and 50 times greater, respectively, than the recommended maximum clinical dose of 0.02 ml/kg for ultrasound imaging). Lung lesions were not observed in cynomolgus monkeys even at the highest dose given (1 ml/kg/day) for 1 month.

- Genotoxicity

Activated Luminity was tested *in vitro* and *in vivo* for potential mutagenicity and clastogenicity. Bacterial mutagenesis (Ames assay), *in vitro* mammalian mutagenesis assay (L5178Y TK +/- mouse lymphoma cell), *in vitro* chromosomal aberration assay (Chinese hamster ovary [CHO] cell), and *in vivo* mouse micronucleus assays were conducted. In addition to these studies genetic toxicity assays (bacterial mutagenesis, *in vitro* chromosome aberration, and *in vivo* rat micronucleus assays) were conducted. Luminity was not genotoxic.

- Carcinogenicity

The sponsor did not conduct carcinogenicity studies.

- Reproduction Toxicity

Fertility

No Luminity related effects on fertility or reproductive performance were observed in rats. Furthermore, there were no Luminity-related effects on uterine or ovarian parameters, or fetal survival at cesarean section on Day 13. Sperm count, morphology and testicular and accessory sex gland weights were also not affected by activated Luminity. There were no histological findings in gonads or other reproductive tissues in 1-month toxicity studies in rats and monkeys.

Embryo-fetal development

Well-characterized activated Luminity was given to pregnant female rats at doses of 0.1, 0.3 or 1.0 ml/kg/day at 1 ml/kg/min from Days 6 to 17 of gestation. Doses were selected based on a range-finding study in which doses ≤ 1.0 ml/kg/day were not associated with maternal toxicity: however, a dose of 3.0 ml/kg/day resulted in maternal death. In the range-finding study, clinical signs prior to death included abnormal respiration, decreased activity and unconsciousness. There was no evidence of activated Luminity-related maternal or developmental toxicity (fetal growth, survival, and morphological development) in the definitive study at ≤ 1 ml/kg/day.

In rabbit studies, Luminity was given to pregnant females at doses of 0.1, 0.3 or 1.0 ml/kg/day at 1 ml/kg/min from Days 7 to 19 of gestation. A dose of 3.0 ml/kg/day resulted in maternal death. Clinical signs prior to death included abnormal respiration, incoordination, decreased activity and/or convulsions. A few maternal deaths occurred at the high dose of 1.0 ml/kg/day and were associated with clinical signs that were similar to those observed during the range-finding study. In addition, transient respiratory signs were observed in the definitive study at doses ≥ 0.3ml/kg/day. There was no evidence of activated Luminity-related developmental toxicity (fetal growth, survival, and morphological development) in this study, even at dose ≥ 0.3 ml/kg/day, which were associated with maternal toxicity.

Pre and Postnatal Development

Pregnant female rats (F_0 generation) were given activated Luminity injected daily at 0.1, 0.3 or 1.0 ml/kg/day from gestation Day 6 to lactation Day 21, 22 or 23. Shortly after dosing, some females exhibited decreased activity and a few deaths (3/22) during the gestation period in the 1.0 ml/kg/day group indicating maternal toxicity. There were no other signs of maternal toxicity and reproductive performance was unaffected. There was no effect on survival, physical development, behavior or reproductive performance of the F_1 generation nor on the survival and *in-utero* development of the F_2 generation. The NOEL for general toxicity (F_0 generation) was 0.3 ml/kg/day. The NOEL for reproductive performance in the dams (F_0 generation), including maintenance of gestation and delivery and nursing of the pups was 1.0 ml/kg/day. The NOEL on growth, development and reproductive performance of the F_0 generation was determined to be 1.0 ml/kg/day.

- Local tolerance

There was no evidence of hemolysis, or vascular or ocular irritation in studies in rabbit. Reversible, mild irritation produced by a single intramuscular injection of 1 ml of activated Luminity was similar to that produced by the weakly acidic positive control, 0.425% acetic acid (pH ~ 2.9). There were no activated Luminity-related gross or microscopic changes at the injection site in the general toxicology studies.

- Other toxicity studies

Antigenicity

The antigenic potential of activated Luminity was investigated in two separate studies in guinea pigs, each involving evaluation of active systemic anaphylaxis (ASA) and passive cutaneous anaphylaxis

(PCA) models. A weak response in the ASA model was not paralleled by a positive result in the PCA model. Similarly, in another study, activated Luminity was not shown to have antigenicity in mice or skin sensitising potential in guinea pigs. Therefore, these studies indicate that there is little potential for immune-mediated hypersensitivity to activated Luminity in humans.

Single Dose Tolerance Study in Cynomolgus Monkeys
In a single intravenous dose study of 3 ml/kg of activated Luminity administered at a rate ~ 1 ml/kg/min to cynomolgus monkeys elicited clinical signs (including abnormal respiration and unresponsiveness) during or immediately following dosing. The absence of any meaningful change in hematology parameters or plasma levels of histamine, tryptase or complement (SC5b-9) in the current study indicated that the acute response to activated Luminity was not an anaphylactoid response mediated by mast cell degranulation or activation of complement system. Abnormal electrocardiographic changes included ST-T segment depression followed by cardiac arrhythmias within 1 minute of initiation of dosing. The acute development of ST-T segment depression is typical of myocardial ischemia [3].
The used dose of 3 ml/kg is 150 times the maximum recommended human dose of 0.02 ml/kg for ultrasound imaging. When this dose is expressed in terms of the rate of dose administration, ~ 1 ml/kg/min, the margin is 50-fold compared to the diagnostic use in humans by bolus injection.

Ecotoxicity/environmental risk assessment

Luminity has been evaluated for potential adverse environmental effects. The focus of this evaluation is the active ingredient perflutren and the phospholipid components. The amount of perflutren that could enter the atmospheric compartment is approximately 10^{-9} lower than the estimated total emissions of greenhouse gases in the EU and as such is not considered to pose a concern. The predicted environmental concentration in surface waters for the phospholipid components is estimated to be less than 2 pg/l. Concentrations below 10 ng/l are not considered to be of concern.

Discussion on the non-clinical aspects

Based on the *in vivo* imaging studies performed in the open chest dog, the potential usefulness of activated Luminity for cardiac imaging has been demonstrated. These findings are consistent with the growing clinical experience.
The preclinical studies indicate activated Luminity is amenable to use for echocardiography. The dose levels to produce optimal images without shadowing in the dog (10 µl/kg) are also consistent with the maximum clinical dose (10 µl/kg single dose with a second 10 µl/kg dose). Collaborative and literature studies with activated Luminity in a variety of animal models (tumor imaging in the rabbit and rat, testicular ischemia in the canine, monitoring placental flow in gravid baboons, blood flow in intraovarian arteries in the pig and myocardial opacification in rhesus monkeys) also support the potential effectiveness of activated Luminity for a range of microcirculation imaging applications.

Pharmacodynamics

The applicant has developed *in-vitro* and used *in-vivo* models to confirm that Luminity is an effective contrast agent to use in conjunction with ultrasound in order to aid in the diagnosis and assessment of myocardial perfusion defects. A correlation demonstrated increased acoustic power and increased pressure will shorten the persistence of microspheres in circulation.
A dose response relationship was found between opacification ability and duration of action. Microspheres between 1 and 10 µm are the primary contributors for imaging in the fundamental mode, whereas microspheres between 1-2 µm contribute primarily to imaging in the second harmonic mode.
An open-chest occlusion/reperfusion model was used to demonstrate *in-vivo* pharmacodynamic effect. Perfusion images demonstrated a correlation between pathological necrotic areas at 20 – 60 minutes, but did not localise the infarct at 2 hrs post reperfusion. Although fundamental imaging was not affected by time post activation up to 12 hours, the loss of microspheres between 1-2 µm resulted in gated second harmonic imaging only being effective at 5 minutes post activation.
No secondary pharmacodynamic studies were performed.

Pharmacokinetics

The amounts of naturally occurring phospholipid (DPPA and DPPC) administered represent an extremely small portion relative to their respective endogenous levels and the metabolic fate of DPPC and DPPA were not followed. Not naturally occurring MPEG5000-DPPE exhibited biphasic clearance with a small volume of distribution and by 72 hours was cleared from the circulation. The MPEG5000-DPPE is hydrolyzed to MPEG 5000- LPE and then slowly cleared via urine as MPEG5000. The rat was chosen for these studies because of its use in toxicity studies and the flexibility of determining tissue distribution in replicate at multiple time points.

The total amount of PFP administered (~350 µl to a 70 kg patient) as part of the activated Luminity clinical dose is low and as a gas with minimal water solubility would be expected to be cleared via the lungs. Studies were therefore conducted to follow blood and expired air levels following activated Luminity administration. The beagle dog was selected for these studies because of the relevance to pre-clinical efficacy and safety pharmacology studies and the established use of dogs in pulmonary expiration studies. PFP expired through the lungs is unchanged.

There is no indication of any long-term retention or potential for accumulation of the non-endogenous components. The rapid elimination of PFP in the expired air is consistent with the rapid disappearance of ultrasound contrast after activated Luminity administration. Since PFP is cleared via the lungs, the possibility exists that PFP could interfere with efficient pulmonary gas exchange and affect P_{O2}. However, the small amount of PFP administered to patients (≤350 µl to a 70 kg patient) compared to the lung capacity (>4 l) and mixing volume (>1 l/min [4] make this unlikely to have any clinical relevance.

Toxicology

The applicant has performed a number of single dose toxicity studies in mouse, rat and dog. Repeat dose toxicity studies were conducted in rat, beagle dog and cynomolgus monkey. The no-observable-effect doses (NOELs) for clinical signs after 1 month of treatment in rats (0.1 ml/kg/day) and monkeys (0.3 ml/kg/day) are 5 and 15 times the recommended maximum clinical dose of 0.02 ml/kg for ultrasound imaging (0.01 ml/kg with possible administration of a second dose of 0.01 ml/kg).
When these no-observable-effect doses are expressed in terms of the rate of dose administered, (2 ml/kg/min in rats and 1.2 ml/kg/min in monkeys), the margin is ≥ 60-fold compared to the injection rate for diagnostic use in humans (0.01 ml/kg administered in 30-60 seconds or 0.01-0.02 ml/kg/min.

Luminity has not shown any mutagenic activity in several in vitro studies carried out in a large range of genotoxicity test. With these data and according to CPMP/ICH/141/95 guideline Luminity can be considered as a non-genotoxic substance.

Genotoxicity studies conducted did not indicate potential of mutagenicity or clastogenicity. Therefore the lack of carcinogenicity studies is considered justified [5].

There is no evidence of Luminity induced embryo-fetal toxicity in rat and rabbit reproductive development studies.

There was no evidence of haemolysis nor vascular or ocular irritation in the local tolerance studies presented.

Luminity does not present an environmental risk following patient use.

4. Clinical aspects

Introduction

The present application is based on a database obtained from 2827 subjects in 39 clinical studies (37 completed and 2 ongoing). All of these studies were included in the evaluation of safety. Three further clinical studies were ongoing at the time of the preparation of this application, but no data were available from those studies at the time of database lock for the safety analyses (30 June 2004). Three of the 37 completed studies provided pharmacology data and 10 contributed to the evaluation of efficacy in echocardiography. Two further studies using non-linear ultrasound imaging (DMP 115-401 and DMP 115-410) have been submitted during the assessment procedure to support the efficacy for the use of Luminity in echocardiography. These studies are described and discussed in the 'Discussion on clinical efficacy'.

GCP

The Clinical trials were performed in accordance with GCP as claimed by the applicant.

The applicant has provided a statement to the effect that clinical trials conducted outside the community were carried out in accordance with the ethical standards of Directive 2001/20/EC.

Pharmacokinetics

Luminity is an ultrasound contrast agent, comprised of phospholipid-encapsulated perfluoropropane (PFP) microspheres, designed to enhance echocardiographic and radiologic ultrasound images. Following intravenous (IV) administration, Luminity creates an echogenic contrast effect in blood, which can be observed during ultrasound imaging. PFP is a component of the approved ultrasound contrast-enhancing agent Optison™. It is an inert gas that is considered to be chemically unreactive and is not metabolised in vivo.

The *in vivo* human pharmacokinetics of Luminity was evaluated in one study (DMP 115-905). The pharmacokinetics and metabolism of the lipid blend (DPPA, DPPC and MPEG5000 DPPE) were not evaluated in clinical pharmacology studies.

Whole blood and expired air were sampled at baseline and at frequent intervals up to 15 minutes following administration of Luminity. PFP concentrations in expired air and blood were determined using the validated gas chromatographic method developed for the preclinical studies.

- Methods

PFP concentrations in blood (encapsulated and free PFP combined) and expired air (free PFP) were measured using a validated gas chromatographic method in order to estimate the pharmacokinetic parameters.

- Absorption

In all subjects, maximal concentrations of PFP (C_{max} range = 0.79-9.52 µl/ml) were achieved at approximately 1 to 2 minutes after start of injection.

- Distribution

Binding of PFP to plasma proteins or partitioning in blood cells has not been studied. However, protein binding is expected to be minimal because PFP has a low partition coefficient into whole blood.

- Elimination

The results in study DMP 115-905 indicated that PFP appeared to be rapidly cleared from the systemic circulation following IV administration of Luminity at the tested dose of 50 µl/kg (CL range: 1,088-4,775 l/h). In most subjects, PFP was undetectable after 4 to 5 minutes in both blood and expired air. PFP blood concentrations were shown to decline in a mono-exponential fashion with a mean half-life of 1.3 minutes in healthy subjects and 1.9 minutes in COPD subjects.

The percent of PFP dose excreted in expired air ranged in all subjects from 15 to 85% (mean ~50%) of the PFP dose administered. On average, in all subjects, lung elimination accounted for approximately 50% of the total systemic clearance for PFP. The apparent low PFP recovery in expired air in this study could be due to the inability to quantify low levels of PFP by gas chromatography coupled with the very low quantities of administered PFP.

- Metabolism

PFP is an inert gas that is considered to be chemically unreactive and is not metabolized in vivo (*Hutter et al., 1999*). No metabolism studies for PFP were performed.

- Dose proportionality and time dependency

Maximal concentrations of PFP (C_{max} 0.79–9.52 µl/ml) were achieved at approximately 1 to 2 minutes after start of injection. This was possibly related to transit from site of administration to site of blood sampling.

In study DMP 115-905, mean time to maximum Doppler intensity (t_{max}) was similar compared to the PFP blood maximum concentration (t_{max}) from the noncompartmental analysis (1.13 vs 1.77 minutes). Though PFP concentrations in blood fell below detectable limits in most subjects by 4 to 5 minutes, the Doppler signal intensity was still measurable at levels well above baseline values. Mean PFP blood half-life for healthy subjects was 1.3 minutes and 1.9 minutes for COPD subjects as estimated from the noncompartmental analysis. The Doppler measurements indicate that microspheres of Luminity containing PFP continue to be present in the circulation for approximately 10 minutes even though the PFP is not detectable by the gas chromatographic assay.

- Special populations

PFP systemic clearance was 26% lower on average in women than men (2907 l/h vs 2302 l/h, all subjects), resulting in an increase of 67% in maximum concentration (C_{max}) and 3% in area under the concentration-time curve extrapolated to infinity (AUC_{∞}). However, none of these differences were statistically significant. CL_{lung} was lower (by 51%) in women than men (all subjects).

The concentration-time curves of blood PFP for both normal subjects and COPD subjects showed a delayed but rapid rise to Cmax for most study subjects. After reaching Cmax, PFP concentrations declined in a log-linear fashion for both normal and COPD subjects. Average concentration-time profiles for all subjects were similar between both groups (normal versus COPD).
No statistically significant differences were observed in the comparison of normal versus COPD subjects for Cmax, tmax, AUClast, AUC t1/2, CL, Vss or CLlung.

- Pharmacokinetic interaction studies

Drug-drug interaction studies have not been performed with Luminity.

Pharmacodynamics

- Mechanism of action

Luminity has no pharmacologic action. However, as confirmed in preclinical studies, Luminity exhibits lower acoustic impedance than blood, thus it produces its effect by increasing the intrinsic backscatter of ultrasound beams from blood. The interface between Luminity bubble and blood acts as a reflector of the ultrasound beam thus enhancing blood echogenicity and increasing contrast between the blood and the surrounding tissues.

- Primary and Secondary pharmacology

Primary pharmacology
DMP 115-900 was a Phase 1 study to determine the safety and tolerability of Luminity given as a single IV bolus injection to healthy adult male subjects. Five ascending doses of Luminity were studied, and each Luminity dose was compared to placebo.
The preliminary efficacy results of DMP 115-900 indicated that the median visual scores for contrast enhancement were higher in the Luminity-treated groups compared with placebo.
Quantitative assessment of the effects of Luminity on the enhancement of the cardiac images was performed on the videodensitometry data. Nearly all mean changes in score from baseline after Luminity were positive, and some were significantly different from placebo. The mean changes were most pronounced for the enhancement of the left ventricle as a whole and the septal and lateral walls, respectively. In conclusion, both the median visual scores for contrast enhancement and the mean changes from baseline in videodensitometry were higher in the Luminity-treated groups than in the placebo-treated group.

DMP 115-901 was a Phase 1 study to determine the safety and tolerability of Luminity given as multiple IV bolus injections in healthy adult male subjects. The preliminary efficacy results of DMP 115-901 indicated that the percentage of subjects demonstrating optimal left-ventricular cavity enhancement during blinded visual evaluation (score of 2 or 3) was higher following the first injection of Luminity than following administration of placebo.
For both ventricles, the magnitude of increase in signal intensity measured by videodensitometry following the first injection was larger for both blinded readers for all Luminity dose groups than for the placebo dose group. Also, the magnitude of increase in signal intensity following the first injection for myocardial wall videodensitometry was generally larger for all Luminity dose groups than the placebo dose group for the anteroseptal and anterior walls, and for the 10 and 15 µL/kg dose groups for the inferoseptal wall.

Secondary pharmacology
The ability of Luminity to elicit an immune response was examined in 128 subjects from four studies. Assessments included immunoglobulins A, E, G, and M, histamine (blood), anti-double stranded deoxyribonucleic acid antibody, tryptase, complement (CH_{50} and C3a), and lupus anticoagulant. No clinically important changes were noted with the exception of activated complement factor C3a. C3a levels increased immediately after Luminity injection then declined over 30 minutes after dosing. The elevated levels had no effect on release of histamine and all subjects tolerated the agent well with no symptoms suggestive of hypersensitivity reaction.

Clinical efficacy

Ten clinical efficacy studies in support of this application for the approval of Luminity in echocardiography have been submitted. Five pivotal Phase III studies (DMP 115-004, 005, 006, 007, and 017) and five supportive Phase 2 studies (DMP 115-018, 022, 209, 211, and 902). All studies were conducted in accordance with good clinical practice (GCP). The five pivotal studies support the use of Luminity for contrast-enhanced echocardiographic imaging of cardiac structure (ventricular chambers and endocardial borders))

Data from two Phase 2 studies (DMP 115-018 and 022) support the use of Luminity in imaging ventricular chambers and endocardial border delineation during stress/rest echocardiography with non-linear imaging in subjects with known or suspected coronary artery disease (CAD).

In addition, supportive data for the determination of the optimal dose of Luminity for fundamental echocardiography are presented from a dose-ranging Phase 2 study (DMP 115-902).

Supportive data presented during the assessment from two further studies (DMP 115-401 and -410) support the claimed indication using non-linear ultrasound technique.

Key Features of the Clinical Studies Supporting the Claims for Efficacy in the Echocardiography Indication

Study	No. Subjects Treated with Luminity (Placebo)	Mode of Luminity Administration	Main Type of Ultrasound Imaging	Type of Control: Parallel Placebo Group	Type of Control: Standard Diagnostic Technique	Type of Control: Unenhanced vs Enhanced Images	Blinded Read
PIVOTAL ECHOCARDIOGRAPHY STUDIES							
DMP 115-004	69 (18)	Bolus	Fundamental	Yes	–	Yes	Yes
DMP 115-005	100 (24)	Bolus	Fundamental	Yes	–	Yes	Yes
DMP 115-006	67 (–)	Bolus	Fundamental	No	MRI	Yes	Yes
DMP 115-007	59 (–)	Bolus	Fundamental	No	MRI	Yes	Yes
DMP 115-017	64 (–)	Bolus + infusion	Fundamental	No	–	Yes	Yes
SUPPORTIVE ECHOCARDIOGRAPHY STUDIES							
DMP 115-018	78 (40)	Infusion	Non-linear	Yes	Nuclear imaging	No	Yes
DMP 115-022	87 (43)	Infusion	Non-linear	Yes	Nuclear imaging	No	Yes
DMP 115-209	69 (–)	Infusion	Non-linear	No	Coronary angiography	No	Yes
DMP 115-211	26 (–)	Bolus + infusion	Non-linear	–*	–*	–*	No
DMP 115-902	42 (14)	Bolus	Fundamental	Yes	–	Yes	Yes

* Study DMP 115-211 was uncontrolled and enrolled healthy subjects.
** The final diagnosis acted as a comparison for diagnostic accuracy; contrast-enhanced CT or MRI was a comparison for concordance
CT=computed tomography, IVP=intravenous pyelography, MRI=magnetic resonance imaging
Note: Institutional reads were performed in all of the 15 studies.

- Dose response study

DMP 115-902 was a single-blind, randomised, multicentre, placebo-controlled, parallel-group, dose-ranging study performed in 29 male and 27 female patients referred for diagnostic echocardiography.

The primary objectives were to determine the safety and tolerability of Luminity as well as to investigate the dose effect of contrast enhancement in facilitating visualisation of the cardiac chambers and enhancement of the myocardial walls following an IV injection of Luminity (42 patients) or a placebo (14 patients) control during a 2-D echocardiographic exam.
56 subjects (mean age was 54.8years) were randomised to receive a single dose of Luminity (5, 10, or 15 µl/kg) or placebo as an IV bolus injection.
Each subject underwent continuous echocardiography using optimal baseline settings starting 3 minutes before administration of study medication and continuing for at least 15 minutes post-injection or until the echocardiographic image returned to baseline.
Images were evaluated by the investigators (institutional read) and by three independent cardiologists who were blinded to subject information, dose of the imaging agent, and type of image (pre-vs post-

injection). All subjects were monitored for safety prior to and following the Luminity or placebo administrations.

Results
The percentages of subjects with optimal enhancement in the Luminity dose groups were as follows:
Reader 1: 5 µl/kg 100.0%, 10 µl/kg 92.3%, 15 µl/kg 100.0%
Reader 2: 5 µl/kg 100.0%, 10 µl/kg 84.6%, 15 µl/kg 100.0%
Reader 3: 5 µl/kg 100.0%, 10 µl/kg 92.3%, 15 µl/kg 100.0%
The percentages of subjects with optimal left-ventricular enhancement were comparable among Luminity dose groups for all readers. Duration of optimal enhancement (blinded read) ranged from 54 to 102 seconds at 5 µl/kg, from 82 to 130 seconds at 10 µl/kg, and from 130 to 179 seconds at 15 µl/kg. Time to optimal enhancement was significantly longer in the 10 and 15 µl/kg groups compared to the 5-µl/kg group.

- Main studies

Efficacy data for the 10µl/kg dose of Luminity are available from the 5 pivotal cardiology trials DMP **115-004, -005, -006, -007, and –017**.

DMP **115-004, -005** are randomized, double-blind, multicenter, placebo-controlled, parallel-group, phase III studies conducted on subjects who had been referred for evaluation of ventricular function and had suboptimal echocardiographic images.

DMP 115-006, -007 are open-label, non-randomized, multicenter Phase III studies conducted on subjects with suboptimal echocardiographic images.

DMP 115-017 is a phase III, multicenter, open-label crossover trial to compare the ability of Luminity, when administered as an infusion versus a slow bolus injection, to visually demonstrate LV cavity enhancement and to improve EBDL in patients with suboptimal echocardiographic images.

Methods

Study Participants

Inclusion Criteria
Studies DMP 115- 004+005 adult patients with suspected or known cardiac disease, referred for evaluation of ventricular function, and pretrial unenhanced suboptimal echocardiographic images in the 4- or 2-chamber view
Studies DMP 115- 006+007 adults, who had a pretrial unenhanced suboptimal echocardiographic images in the 4- or 2-chamber view (suboptimal is considered if at least two of six segments of the ventricular border were not evaluable)
Study DMP 115-017 adults, with suspected cardiac disese, who had a pretrial unenhanced suboptimal echocardiographic images in the 4-chamber view

Exclusion Criteria
Patients who met any one of the following criteria were excluded from the trials:
-Presented with a contraindication relative to MRI imaging (e.g., indwelling pacemaker, intracranial clips, severe claustrophobia, intra-auricular or intra-ocular implants, metal fragments in the eyes, etc.)
-Had a history of acute disease that could influence the patient's ability to complete trial procedures (unstable angina, acute myocardial infarction [within 90 days], marked ventricular ectopy [>10 premature ventricular contractions per minute], atrial fibrillation with variable ventricular response)
-Had existing NYHA Class IV CHF or severe COPD
-Had an existing right-to-left ventricular shunt

Treatments

Studies DMP 115-004 and -005

Patients were randomly assigned to one of three dose groups (placebo, 5 µ/kg Luminity, or 10 µl/kg Luminity). Patients in each dose group were assigned to receive two identical single IV bolus dose administrations in a large forearm vein followed immediately by a 10-ml saline flush at a rate of 1 ml/sec. The two dose administrations were separated by a minimum 30-minute period.

Echocardiographic imaging sessions began about 60 seconds before each dose and continued for about 5 minutes after the injection. On the first session, images were acquired for 30 seconds each at the apical 4- and 2-chamber views, both at baseline and postinjection. Following at least a 30-minute period, there was a second imaging session with imaging acquisition for at least 10 seconds per view (apical 4- and 2-chamber; parasternal long-axis; subcostal 4-chamber; and mid-ventricular, apical, and basal short-axis), both at baseline and postinjection.

Studies DMP 115-006 and -007
All patients were scheduled to receive two 10 µl/kg IV injections of Luminity. Each dose was administered through an 18- to 20-gauge needle situated in a large forearm vein. There was a minimum 20-minute washout period between the injection for Imaging Session 1 and the injection for Imaging Session 2.
Fundamental 2-D gray-scale images were obtained before and after the first Luminity injection beginning with the acquisition of pre-contrast baseline images. Image acquisition for each session began approximately 2 minutes before each administration of the trial medication (Luminity) and continued for approximately 60 seconds (20 seconds at each of the following myocardial views: apical 4-chamber view, apical 2-chamber view, and parasternal mid-ventricular short-axis view) following dose administration.

Study DMP 115-017
All patients underwent two imaging sessions (A and B) with Luminity; the imaging sessions were separated by 24 to 72 hours. During imaging session A, each patient received an infusion of 1.3 ml of Luminity diluted in 50 ml of preservative-free saline. During imaging session B, each patient received two single bolus injections of 10 ml/kg of Luminity (total dose during session B was 20 ml/kg). The injection was given as a slow bolus over 30 to 60 seconds, followed by a 5 ml saline flush administered over two minutes.
Echocardiographic imaging of the apical 4-chamber view was obtained prior to, during, and following each of the imaging sessions.

Objectives

DMP115 –004 –005 -017
The primary objectives of these trials were to assess :

- The ability of Luminity to visually demonstrate LV cavity enhancement
- The ability of Luminity to improve endocardial border delineation
- The safety of two single IV doses of Luminity.

The secondary objectives of these trials were to evaluate or examine:

- Duration of LV cavity enhancement
- The ability of contrast enhancement provided by Luminity to improve the diagnostic confidence, wall-motion detection and quality of EF determination.

DMP115 –006 –007
The primary objective of these trials were to compare the accuracy of echocardiographic ejection fraction (EF) measures obtained before and after the administration of Luminity relative to EF assessments determined from magnetic resonance imaging (MRI).

The secondary objectives of this trial were as follows:

- evaluate the number of patients who show improvement in the number of ventricular segments correctly characterized after Luminity administration relative to MRI during wall-motion evaluation

- examine the percentage of patients for whom EF classification changes and for whom EF classification is correctly identified after Luminity administration relative to MRI

Outcomes/endpoints

Results for the efficacy of the bolus dose reported in this summary are based on the first 10µL/kg bolus injection only. A second bolus dose was routinely given in all studies, as this is likely to occur in clinical practice; it was primarily included to demonstrate safety.

In studies DMP **115-004, 005, and 017** the primary efficacy variable was the left-ventricular cavity enhancement score based on the pair wise qualitative interpretation of the first baseline image and the post-injection image for each subject.
Patients were classified according to one of the two following categories:
"Enhanced" corresponding to a score of 2 or 3 (adequate or full enhancement)
"Other" corresponding to a score of 0, 1, or 9 (no, weak, or excessive enhancement)

The primary objective of studies DMP **115-006 and 007** was to compare the accuracy of echocardiographic ejection fraction measures obtained before and after the administration of DMP 11 Luminity to ejection fraction measures obtained from MRI.

$$\frac{100*\left|[\text{EF (Echo without contrast)-EF (MRI)}]\right|}{\text{EF (MRI)}} - \frac{100*\left|[\text{EF (Echo with contrast)-EF (MRI)}]\right|}{\text{EF (MRI)}}$$

All studies Endocardial border delineation was measured in all of the 5 pivotal studies. An improvement in delineation occurred when a segment that was scored as non-evaluable at baseline was scored as evaluable in the corresponding apical view after injection with Luminity. In studies DMP 115-004, 005, 006, and 007, the evaluation was based on the 12-segment model for the apical 4- and 2-chamber views. In study DMP 115-017, the evaluation was based on the six segments visualized in the apical 4-chamber view.

Secondary efficacy measures were Wall motion also evaluated as a secondary objective in each of 12 myocardial segments according to a five-point scale (0=non-evaluable, 1= normal or hyperkinetic, 2=hypokinetic, 3=akinetic, 4=dyskinetic). The number of myocardial segments with an exact match in wall-motion grade between MRI and echocardiography with and without Luminity was determined. The number was converted to a percentage ([number matching segments/12] *100) to measure agreement between the echocardiographic and MRI evaluations. An analogous evaluation based on a classification of wall motion as normal (score: 0 or 1) or abnormal (score: 2, 3, or 4) was also performed.

Sample size

-004 –005 The sample size was calculated for 80% power and a two-tailed test for significance level of 0.05. A sample size of 31 patients per dose group was necessary to show a difference in the duration of contrast enhancement between the 5 and the 10 µL/kg doses. When a 2:2:1 ratio (Luminity 10 µl/kg: Luminity 5 µl/kg:placebo) was used, 80 patients (32:32:16) were needed to be enrolled in this trial.

-006 –007 -017 Assuming a Type I error of 0.05 and 80% power (Type II error of 0.2), a sample size of 50 was needed for the 10% expected mean difference in relative error from MRI in EF.

Blinding (masking)

DMP115-004 -005 were double-blinded placebo controlled trials.

DMP115-006 -007 -017 were open-label trials; therefore, no methods were needed to blind the trial medication. Image evaluation was performed via a blinded evaluation of the imaging tapes, independent of imaging session knowledge.

Statistical methods

-004 -005 A two-sample test of proportions, with Fisher's exact test was used. Statistical significance was determined at the 0.025 level, with Bonferroni adjustment.

-006 -007 For the primary efficacy analysis, a paired t-test was applied to the mean difference in relative error from MRI in EF measured by fundamental echocardiography using data from the blinded read.

Results

Participant flow / Numbers analysed / Recruitment

Study 115-004
The protocol specifies to enroll 80 patients across nine sites into this trial, randomly assigned to each dose group was: 16 for placebo and 32 for each of the DMP-treated groups (5 and 10 µL/kg). Of the 87 patients who were randomized and entered into the trial, 86 patients (98.9%) completed the trial. Any patient who did not receive the second dose of Luminity or placebo was excluded from the analysis of myocardial views with contrast enhancement, since this assessment was only conducted following the second injection.

Study 115-005
There were 124 patients enrolled into this trial across nine sites. Of the 124 patients who were randomized and entered into the trial, 122 patients (98.4%) completed the trial. Two subjects in the 10 µl/kg Luminity group did not complete the study (withdrawn after the first injection due to AEs).

Study 115-006
50 patients were expected to enter. Overall, 67 patients from 4 sites were enrolled in the trial; of those, 66 (99%) completed the trial. The one patient (Patient 3/8) who did not complete the trial had two episodes of dizziness within 10 minutes after the first injection of Luminity and, consequently, did not receive the second injection.
MRI evaluations of EF were not available for four patients because of extremely poor image quality, which prohibited interpretation. Therefore, these four patients were not included in the primary efficacy analysis (or as applicable in secondary efficacy analyses).
61 patients had images obtained with harmonic techniques.

Study 115-007
50 patients were expected to enter the trial. Overall, 59 patients from 4 sites were enrolled in the trial; all patients completed the trial.

Study 115-017
Up to four sites were targeted to enroll 15 to 20 patients each. Up to four sites were targeted to enroll 15 to 20 patients (no site could enroll more than 30 patients). On treatment Day 1, patients were randomized to undergo either imaging session A or B. On treatment Day 2, patients received the alternative imaging session.
Three patients were excluded from the primary efficacy analysis because they did not complete both imaging sessions. Therefore, 61 patients were included in the primary analysis for which direct comparison of bolus and infusion administration results were required.

Conduct of the study

Study 115-004
There were no changes in the planned conduct of the trial. 4 patients failed to have all required ECGs and only 10 patients failed to have all five laboratory profiles completed through 72 hours.

Study 115-005
One hundred ten patients (89%) had at least one protocol deviation. 9 patients failed to have all required ECGs and only 12 patients failed to have all five laboratory profiles completed through 72 hours.

Study 115-006
Fifty patients had at least one protocol deviation. Overall, the deviations were minor and had no effect on trial results (e.g., 34 of the 41 early or late ECGs were obtained within 1 hour of the protocol specified time).

Study 115-007
Forty-four patients had at least one protocol deviation. Overall, the deviations were minor and had no effect on trial results (e.g., 31 of the 42 early or late ECGs were obtained within 1 hour of the protocol specified time).

Study 115-017
All sixty-four patients had at least one protocol deviation. Among deviations, the most frequent were of vital signs (58 patients) or ECGs (60 patients) not obtained within the protocol-specified time windows (e.g. within 30 minutes). Overall, the deviations were minor and had no effect on trial results.

Baseline data

Study 115-004
A total of 87 subjects were enrolled and treated: 69 (79.3%) were male and 18 (20.7%) were female; The overall mean (standard deviation [SD]) age was 62.5 (12.3) years. The three dose groups were similar with respect to age, weight, race, and gender. 10 (11.5%) patients had a history of COPD and 33 (37.9%) patients had a history of CHF. 83 (95.4%) patients were taking cardiovascular medication and 69 (79.3%) patients were taking anticoagulants.

Study 115-005
A total of 124 subjects were enrolled and treated: 70 (56.5%) were male and 54 (43.5%) were female; The overall mean (SD) age was 52.1 (16.3) years. The three dose groups were similar with respect to age, weight, race, and gender.
5 (4.0%) patients had a history of COPD and 29 (23.4%) patients had a history of CHF. 69 (55.6%) patients were taking cardiovascular medication and 50 (40.3%) patients were taking anticoagulants.

Study 115-006

Men and women comprised 52.2% (35 of 67) and 47.8% (32 of 67) of the population, respectively. The mean (SD) age was 48.9 (16.8) years. Overall, 3 (4.5%) patients had a history of COPD, and 11 (16.4%) patients had a history of CHF. 32 (47.8%) patients used cardiovascular medications.

Study 115-007

Men and women comprised 67.8% (40 of 59) and 32.2% (19 of 59) of the population, respectively. The mean (SD) age was 55.3 (13.9) years. Overall, 5 (8.5%) patients had a history of COPD, and 21 (35.6%) patients had a history of CHF. The most common illnesses were heart and lung findings in 19 (32.2%) patients. 49 (83.1%) patients used cardiovascular medications.

Study 115-017
A total of 64 subjects were enrolled and treated: 45 (70.3%) were male and 19 (29.7%) were female. The mean (SD) age was 60.9 (15.7) years. Overall, the most common finding was a history of cardiovascular illness or injury in 47 (73.4%) patients. The most common findings were

cardiovascular and pulmonary in 30 (46.9%) patients. 43 (67.2%) patients used cardiovascular medications.

Outcomes and estimation

Adequate or Full Left-Ventricular Cavity Enhancement

DMP 115-004 and 005 The table below presents the percentage of subjects with adequate or full left-ventricular cavity enhancement based on the blinded read results in the qualifying view. In each study, the results from all three blinded reads indicated that the percentages of subjects who demonstrated adequate or full left-ventricular cavity enhancement after the injection of 10 µl/kg of Luminity were significantly greater than the percentages achieved after injection with placebo. The median percentage of subjects with ventricular enhancement was similar for the blinded reads in the two studies: 60.6% in study DMP 115-004 and 61.2% in study DMP 115-005.

Following administration of 10 µl/kg of Luminity in trials DMP 115-004 and -005 combined, the mean duration of ventricular enhancement was approximately four minutes and the mean duration of clinically useful cavity enhancement was approximately 1.5 minutes.

In study **DMP 115-017**, the results for the three blinded readers indicated that 86% to 98% of subjects demonstrated adequate or full ventricular cavity enhancement for the qualifying view (apical 4-chamber) for the first 10 µl/kg bolus injection of Luminity. By contrast, the blinded readers determined that no subjects (0%) demonstrated adequate or full ventricular cavity enhancement at baseline. The percentages of subjects with adequate or full enhancement of images following bolus administration of Luminity were significantly higher than baseline based on the comparison of confidence intervals.

In trial DMP 115-017 the mean duration of clinically useful ventricular cavity enhancement following the first bolus injection as evaluated in a blinded read ranged from 155 to 234 seconds for the 64 patients evaluated.

Percentage of Subjects with Adequate or Full Left-Ventricular Cavity Enhancement for the Blinded Read Based on the Subjects' Qualifying View in Studies DMP 115-004, 005, and 017 (Echocardiography)

DMP 115-004	Reader 1			Reader 2			Reader 3		
Dose Group	N	% Subjects Enhanced	(95% CI)	N	% Subjects Enhanced	(95% CI)	N	% Subjects Enhanced	(95% CI)
Placebo	18	0.0	(0.0, 17.3)	18	0.0	(0.0, 17.3)	18	0.0	(0.0, 17.3)
Luminity	33	60.6*	(42.2, 73.9)	33	63.6*	(45.1, 76.3)	33	60.6*	(42.2, 73.9)
DMP 115-005	Reader 4			Reader 5			Reader 6		
Dose Group	N	% Subjects Enhanced	(95% CI)	N	% Subjects Enhanced	(95% CI)	N	% Subjects Enhanced	(95% CI)
Placebo	24	0.0	(0.0, 13.6)	24	0.0	(0.0, 13.6)	24	0.0	(0.0, 13.6)
Luminity	49	42.9*	(29.1, 55.8)	49	77.6*	(63.0, 85.9)	49	61.2*	(46.2, 72.6)
DMP 115-017	Reader 1			Reader 2			Reader 3		
Dose Group	N	% Subjects Enhanced	(95% CI)	N	% Subjects Enhanced	(95% CI)	N	% Subjects Enhanced	(95% CI)
Luminity	64	96.9	(88.2, 98.0)	64	85.9	(74.5, 91.5)	64	98.4	(90.5, 98.4)

* Indicates statistically significant difference from placebo, $p \leq 0.01$, in studies DMP 115-004 and 005, only. In study DMP 115-017, the percentages of subjects with adequate or full enhancement of images following bolus administration of Luminity were significantly higher than baseline based on comparison of the CIs.

Note: Luminity dose=First 10 µl/kg bolus injection.

Endocardial Border Delineation (EBDL)

EBDL was assessed as the number of segments that changed from non-evaluable at baseline to evaluable post injection of 10 µl/kg Luminity.

A positive net percentage indicated that more segments changed from non-evaluable at baseline to evaluable post injection than changed from evaluable at baseline to non-evaluable post injection.

Endocardial Border Delineation - Net Percentage of Segments with Change in Evaluability for the Blinded Read in Studies DMP 115-004, 005, 006, and 007

DMP 115-004	Reader 1			Reader 2			Reader 3		
Dose Group	N	Mean (SD)	(95% CI)	N	Mean (SD)	(95% CI)	N	Mean (SD)	(95% CI)
Placebo	18	0.0 (14.3)	(–7.1, 7.1)	18	–4.2 (15.7)	(–12.0, 3.7)	18	–1.9 (13.0)	(–8.3, 4.6)
Luminity	33	24.0*# (25.7)	(14.9, 33.1)	33	2.3 (22.4)	(–5.7, 10.2)	32	9.4# (18.8)	(2.6, 16.1)
DMP 115-005	Reader 4			Reader 5			Reader 6		
Dose Group	N	Mean (SD)	(95% CI)	N	Mean (SD)	(95% CI)	N	Mean (SD)	(95% CI)
Placebo	24	–1.4 (20.	(–9.9, 7.1)	24	4.9 (26.7)	(–6.4, 16.1)	24	4.9 (17.2)	(–2.4, 12.1)
Luminity	49	13.8*# (28.	(5.7, 21.8)	49	13.1# (31.4)	(4.1, 22.1)	49	13.4# (30.5)	(4.7, 22.2)
DMP 115-006	Reader 1			Reader 2			Reader 5		
Read Type	N	Mean (SD)	(95% CI)	N	Mean (SD)	(95% CI)	N	Mean (SD)	(95% CI)
Unpaired	67	20.9# (18.5)	(16.4, 25.4)	67	25.2#(24.5)	(19.3, 31.2)	67	49.6# (22.6)	(44.1, 55.1)
Paired	67	22.8# (19.3)	(18.0, 27.5)	67	41.7#(24.7)	(35.7, 47.7)	67	54.1# (22.9)	(48.5, 59.7)
DMP 115-007	Reader 3			Reader 4			Reader 5		
Read Type	N	Mean (SD)	(95% CI)	N	Mean (SD)	(95% CI)	N	Mean (SD)	(95% CI)
Unpaired	59	0.7 (19.3)	(–4.3, 5.7)	59	17.7#(33.6)	(8.9, 26.4)	59	41.8# (23.4)	(35.7, 47.9)
Paired	59	6.5# (19.8)	(1.3, 11.6)	59	29.4#(24.9)	(22.9, 35.9)	59	48.3# (25.3)	(41.7, 54.9)

* Indicates significant difference from placebo, p≤0.05

# Indicates significant change in evaluability, p≤0.05

Note: Luminity dose=10 µl/kg. Efficacy results for studies DMP 115-006 and 007 are based on fundamental echocardiography injection 1.

In trials DMP **115-004 and -005**, in five of the six blinded readers, Luminity administration resulted in a significant improvement in endocardial border delineation over the non contrast enhanced baseline

echocardiography examination. Results were less marked for the apical 2-chamber view. Results for two of the six blinded reads were statistically significantly greater for Luminity than placebo for the net percentage of segments with *changes in the evaluability*.

In trials DMP **115-006 and -007**, images were presented in both unpaired and paired format during the blinded read. For patients receiving 10 µl/kg of Luminity in trials DMP 115-006 and -007 statistically significant positive net percentages were seen for five of the six unpaired blinded reads and for all of the six paired blinded reads.
For the unpaired blinded reads the median values for net percentage of segment improvement were 25.2 and 17.7 per cent for trials DMP 115-006 and -007 respectively. For the paired blinded reads the median value for net percentage of segment improvement in these two trials were 41.7% and 29.4% respectively.

In trial **DMP 115 -017** there were significant positive mean net percentages seen for all three blinded reads.

For trials DMP **115-004 and -005**, the percentages of patients who showed improvements in endocardial border delineation **in at least one segment** were greater in patients receiving 10 µL/kg of Luminity *compared to placebo* for all six blinded reads although the equivalent results for placebo in -004 were 35-44% and in -005 54-67%. There were similar results in trials DMP 115-006, -007 and -017.
For percentages of patients showing improvement in at least two segments the majority of numbers suggested improvement **in at least two segments** on the post contrast examination in each of the five echocardiography for the blinded reads. Equivalent figures for the placebo group in -004 were 17-23% and –005, 23-55%.

- **Ancillary analyses**

Study 115-004 -005
The percentage of patients demonstrating adequate or full LV cavity enhancement was 0% for the placebo group for all subgroups and was generally much higher for the 5 and 10 µl/kg dose groups for all subgroups. The results were consistent within each subgroup (gender, age, race, history of COPD, history of CHF).

Study 115-006 -007
Difference in relative error from MRI in EF showed that none of the subpopulations (except for COPD) achieved a 10% or greater improvement. Overall, results showed that Luminity converted more segments from non-evaluable to evaluable than from evaluable to non-evaluable (relative to baseline) for all subpopulations, and the results were consistent between subgroups for subpopulation.

Study 115-017
The results of both subpopulations by gender and age (<65 versus ≥65) were consistent with the primary efficacy results

- Analysis performed across trials (pooled analyses and meta-analysis)

Analyses of results from studies that had similar or identical protocols are presented together.

- Supportive studies

A total of five supportive echocardiography studies were performed: DMP 115-018, 022, 209, 211 and 902. All studies except DMP 115-902 used non-linear ultrasound imaging.

DMP 115-018 and DMP 115-022 were open-label, randomised, multicentre, placebo-controlled, parallel-group conducted in subjects who were under evaluation for suspected CAD. Subjects with screening echocardiographic images that demonstrated a resting wall

motion abnormality were to be excluded. Subjects in study DMP 115-022 had to have screening echocardiograms but there was no requirement that they should be difficult to interpret.
The **primary efficacy** measure was the diagnostic accuracy of echocardiographic images in the detection of CAD**.**
Both studies (DMP 115-018 and DMP 115-022) failed primary efficacy analysis which was based on the comparison of the ability of at least two blinded readers to detect exercise stress-induced wall motion abnormalities from echocardiography and classification of CAD based on angiographic findings. In general sensitivity was low.
In DMP 115-018, the sensitivity to detect CAD relative to nuclear perfusion was quite low among the blinded readers (5.3,10.5 and 26.3) following enhancement. Reader 3 had a statistically significant difference from placebo but reader 3 sensitivity for placebo was 0. Agreement in DMP group (50-58) was lower than in the placebo group (57-60).
In DMP 115-022, the sensitivity was low among the blinded readers (32.1, **39.3** and 25) following enhancement. For the placebo group equivalent figures were 15.4, **53.8** and 0. Sensitivity therefore reduced for reader 5 (53.8 with placebo and 39.3 with enhancement). Agreement in DMP group (63-73) was slightly higher than in the placebo group (58-67).
The studies also failed in Detection of Inducible Ischaemia, Detection of Resting Abnormalities and Diagnostic Accuracy Based on Final Diagnosis.

The studies were successful in a post hoc analysis. Wall Segment Evaluability, which was not planned in the protocol, was performed by considering number of evaluable segments per subject and by per segment analysis. Luminity contrast-enhanced echocardiography resulted in a statistically significantly greater mean number of evaluable segments per subject at rest in the Luminity treatment group compared to the placebo group ($p<0.001$). This treatment improvement was maintained during exercise stress.

DMP 115-209 was an open-label, non-randomised, Phase 2, multicentre study in which the presence or absence of CAD based on the blinded read of the echocardiographic images was compared with the institutional results of the angiographic images following dipyridamole. Patients had undergone or were going to undergo coronary angiography within 30 days prior or 90 days after enhanced echo imaging.

The primary objective was to evaluate the ability of Luminity -enhanced echocardiographic imaging to detect inducible (during dipyridamole stress) perfusion abnormalities in subjects with known or suspected CAD, as confirmed by coronary angiography. The secondary objectives were to assess the safety of Luminity during dipyridamole stress and to evaluate the diagnostic accuracy of DMP-enhanced echocardiographic imaging as compared with coronary angiographic confirmation of disease to localise inducible perfusion abnormalities by vascular territory. Imaging was performed with low mechanical index (MI), real-time perfusion techniques.

Blinded read of Luminity -enhanced echocardiography results demonstrated 86% sensitivity and 80% specificity for the detection of CAD using coronary angiography as the truth standard. A similar sensitivity (88%) was demonstrated for myocardial perfusion in 11 subjects using Luminity administered at the IV infusion dose labelled in the USA for left-ventricular opacification and endocardial border delineation.

A subset of patients from the original protocol was selected for blinded read on the basis of use of Philips software and hard ware upgrade and this reduced the patient numbers from 58 to 41. Consensus reading was not possible in 10 of the 41 cases and so sensitivity and specificity were based on relatively small numbers (31 angiography and 11 in the infusion group).

Study DMP 115-211 was an open-label, non-randomised, single-centre, crossover Phase 2 study conducted on healthy subjects with no evidence of cardiovascular disease. The primary objective was to determine the optimal IV dose and delivery technique (bolus injection versus neat infusion) of Luminity for assessing myocardial contrast echocardiography MCE and the primary efficacy measure was a quantitative segmental analysis of MCE. MCE was assessed by applying a four-point scoring system

to a 12-segment echocardiography model: 0 = minimal or no MCE, 1 = patchy MCE, 2 = homogenous (complete) MCE, a = artifact. Successful MCE was defined by a score of 1 or 2. Myocardial perfusion image acquisition was performed using two low-MI pulse-inversion Doppler acquisition techniques: continuous imaging and 1 to 1 ECG-gated imaging.

As the dose of Luminity increased, independent of bolus injection or infusion dosing method, the percent of successful MCE increased, thus demonstrating a dose response. Luminity was equally effective in producing successful MCE when delivered by either bolus injection or infusion. The first 20 subjects received neat infusions and bolus injections on independent days, and Luminity produced consistent and reproducible MCE on both days ($p<0.001$). Low-MI, ECG-gated imaging resulted in a greater degree of MCE than the continuous low MI imaging technique independent of dosing method. The data suggests that reapplication of a bolus dose before each view would further improve MCE.

The blinded read results for the primary efficacy variable were similar for the two groups of subjects: neat infusion 86% sensitivity (95% CI: 73.4%, 98.0%), dilute infusion 88% sensitivity (95% CI: 67.0%, 108.0%). Although the sample size of the subjects who received the dilute infusion was small, the results suggested that Luminity was equally effective when given as a neat IV infusion or as an infusion of 1.3 to 2.6 ml diluted in 50 to 100 ml saline.

- Discussion on clinical efficacy

The demonstration of efficacy in the initial MAA to support the use of Luminity in Echocardiography contained 10 studies, 5 pivotal and 5 supportive studies of which 4 used the non-linear ultrasound technique.

In three of the pivotal studies using harmonic imaging, the primary variable related to intensity of left-ventricular cavity enhancement (judged in a subjective but blinded way) being shown that the percentage of subjects with “optimal” intensity was higher in the contrast than in non-contrast (placebo or no injection) groups in a population of subjects where conventional non-contrast echocardiography was considered sub-optimal using pre-established definitions.

Two of the main secondary variables studied in the pivotal trials were endocardial border delineation (EDBL) and wall motion. For EDBL Luminity increases the number of evaluable segments and in some subjects with non-diagnostic (i.e. non-assessable) echocardiograms (four or more non-evaluable segments in a single apical view) Luminity resulted in diagnostic (i.e. assessable but not necessarily correct) echocardiograms. As for ‘wall motion’, including after ‘stress’ (critical for the diagnosis of coronary artery disease), there was an increase after Luminity in individual segments identification.

Studies DMP 115-018 and -022 have demonstrated efficacy in a post hoc analysis in Wall Segment Evaluability defined by considering number of evaluable segments per subject and by per segment analysis.

The CHMP consulted the Diagnostics Scientific Advisory Group about the possibility that results with Luminity in fundamental imaging can be extrapolated to non-linear imaging techniques. The advisory group concluded that non-linear imaging is the present state of the art for unenhanced as well as for enhanced ultrasound imaging. In general, it is not possible to assume that the effect of a contrast agent in fundamental imaging will be the same using non-linear imaging.
Following the consultation of the expert meeting and in response to the major objections, the applicant submitted preliminary reports from two further studies DMP 115-410 using harmonic and DMP 115-401 using non-linear imaging to support the claimed indication in echocardiography.

Study DMP 115-401 was an open-label, randomised Phase IV multicentre trial that included 560 patients from 27 sites. The study was designed to determine whether the use of Luminity (DMP115) during stress echocardiography decreases the need for additional diagnostic testing and the number of non-diagnostic imaging studies. Patients were required to have baseline echocardiographs classified as difficult-to-interpret (i.e. 2 or more consecutive segments not adequately visualised) and were randomised to either Luminity enhanced rest and stress imaging or unenhanced rest and stress

imaging. Following imaging, the investigator determined if another diagnostic test was required and at 90 days, a review was undertaken to determine whether additional diagnostic testing had been performed. 376 patients received Luminity and 184 patients underwent unenhanced imaging.
Results show that the use of Luminity contrast material to enhance echocardiography both at rest and during stress significantly improved diagnostic results over unenhanced testing. A smaller percentage of subjects in the Luminity treatment group (12.2%) were recommended a follow-up diagnostic test than in those subjects who had unenhanced echocardiography (32.6%). The difference was highly statistically significant ($p<0.0001$) according to the Fisher exact test.

Investigator confidence was improved in 78.7% of subjects who received Luminity compared with 3.3% of subjects who had unenhanced echocardiographic imaging at rest. Echocardiographic imaging after an exercise stress test showed Investigator confidence was improved in 83.2% of subjects who received Luminity compared with 9.8% of subjects who had unenhanced echocardiographic imaging. Differences between treatment groups were highly significant according to the Wilcoxon rank sum test; p-values were <0.0001 for imaging both at rest and after the exercise stress test.
Significantly fewer tests were actually performed in the Luminity-enhanced group (17%) than in the unenhanced group (36%), a statistically significant difference ($p<0.0001$). Despite the reduction in number of tests, outcomes in the two groups were similar at 3, 6, and 12 months, indicating that diagnoses had not been missed.

Study DMP 115-410 was an open-label, phase IV study conducted at 4 study sites in the USA. The primary objective of this study was to test the hypothesis that Luminity improves the diagnostic accuracy of dobutamine stress echocardiography in detecting and evaluating the extent of coronary artery disease (CAD) compared to non-contrast imaging, using coronary angiography as the standard for comparison.

A total of 100 subjects with an intermediate to high probability of CAD were to undergo 2 dobutamine stress echocardiograms, 1 with Luminity (i.e. enhanced.) and 1 without Luminity (i.e. unenhanced). The investigator randomized the order of each subject's echocardiograms, and a 4 to 72 hours break separated the two procedures.

Baseline images were obtained in the standard apical and parasternal views before performing stress echocardiography.

The dobutamine infusion was started at 5 µg/kg/min. The dose was increased to 10, 20, and 40 µg/kg/min at 3 minute intervals until the target heart rate was achieved.

Intravenous boluses of 1 to 2 ml of Luminity solution were given at rest and at 3 stages of the dobutamine infusion (5 µg/kg/min, 10 µg/kg/min, and at peak dose) so that all digitized images were contrast enhanced.

All subjects were to have had quantitative coronary angiography performed within 60 days prior to or following the dobutamine stress echocardiograms. Angiography was performed in multiple projections using the conventional Judkins technique. Subjects with myocardial infarction or revascularization procedures performed between the echocardiographic studies and coronary angiography were to be excluded. Coronary angiograms were evaluated by an independent observer and diagnosis of CAD was based on the degree of coronary artery stenosis.
Echocardiography was performed using harmonic imaging techniques. Echocardiograms were evaluated by 3 independent readers who were blinded to the clinical information during the reads.

Comparisons of the performance of Luminity-enhanced echocardiography and unenhanced echocardiography were performed. Two measures were assessed by echocardiography, presence of ischemia and presence of abnormality, and compared with presence of disease as diagnosed by angiography. For angiography, presence of disease was classified as having a stenosis $\geq 50\%$ or $\geq 70\%$. In the case of subjects where only enhanced echocardiography allowed an assessment, unenhanced echocardiography was treated as a failure in the agreement analysis. The percent of

evaluable segments for each subject was summarized and compared using a t-test to evaluate whether the mean percent of evaluable segments was the same in each group.

108 subjects were enrolled and 105 completed the study. The reasons for discontinuation in the 3 subjects who did not complete the study were withdrawal of consent in two subjects and 'other' in one subject

Luminity contrast enhancement significantly increased the mean (±SD) overall percentage of segments with adequate visualization from 72 (±24%) to 95 (±8%) at baseline and from 67 (±28%) to 96 (±7%) at peak dobutamine stress ($p<0.0001$). Similar increases were seen in subjects with and without CAD as defined by ≥50% stenosis in angiography.
Unenhanced echocardiograms were uninterpretable in 8 subjects. All of these subjects had interpretable echocardiograms during Luminity-enhanced imaging.
Overall, the readers. confidence of interpretation was assessed as high in a much larger proportion of subjects during Luminity-enhanced echocardiography than during unenhanced echocardiography: 74% (75/101) vs 36% (38/105) subjects.

No studies with similar licensed comparator products have been provided. The Applicant committed to perform a post approval study with the objective to demonstrate the non-inferiority of Luminity versus SonoVue to produce left ventricular cavity opacification and complete endocardial border delineation during exercise stress echocardiography.

Clinical safety

Forty-two clinical studies with Luminity had been performed or were ongoing at the time when this application was prepared.

Data from 37 completed studies performed with Luminity were available.
In addition to the 10 echocardiography studies and 5 radiology studies used to support the claims of efficacy made in this application, the evaluation of safety also reviews safety data from 22 other completed studies: 3 clinical pharmacology studies, 15 echocardiography studies, and 4 radiology studies.

- Patient exposure

Number of Subjects Exposed to Luminity in Echocardiography and Radiology Studies, by Indication and Category of Study

Study Category	Echocardiography	Radiology	Combined
Pivotal	359	309	668
Supportive	302	94	396
Other	897	565	1462
Combined	1558	968	2526

In *echocardiography* studies, about one third of all subjects treated with Luminity received the agent as bolus injections and two-thirds as infusions. Overall, 385 (70.4%) of the 547 echocardiography subjects who received a bolus injection of Luminity received the agent in a dosage of 10-20 µL/kg.

In *radiology* studies, about two thirds of the Luminity-treated subjects received the agent as a bolus injection and the remainder as an infusion. Pivotal radiology studies DMP 115-009 and 010 were performed using bolus injections of Luminity in 209 liver and/or kidney subjects, while the later pivotal radiology study DMP 115-013 investigated the administration of Luminity as an IV bolus and/or infusion in 100 liver subjects.

- Adverse events

Adverse Events in Healthy Subjects
The overall incidence of new-onset AEs in Luminity-treated healthy subjects was 25.0% (11 of 44 subjects). Treatment-related new-onset AEs occurred in 5 (11.4%) of the 44 Luminity-treated subjects. The most frequently reported new-onset AEs in Luminity-treated healthy subjects were headache (3 of 44 subjects), nausea (2 subjects) and flushing (2 subjects). Nausea and flushing were the most frequently reported treatment-related new-onset AEs (two subjects each).

Adverse Events in Clinical Studies
Of the 1899 individuals who participated in the clinical trials, 1716 patients were treated with Luminity. Based on the number of exposures, the most frequently occurring treatment-related adverse events (AEs) were headache (2.3%), back/renal pain (1.2%), flushing (1.1%), and nausea (1.0%).

The undesirable effects reported with Luminity were, in general, non-serious, transient and resolved spontaneously without residual effects.

The adverse reactions observed in more than 1700 adult patients in clinical trials are:

Occurring in ≥ 0.5% of all subjects:

Body as a Whole:	application site disorders, injection site reactions, back pain, chest pain
Digestive System:	nausea
Nervous System:	headache, dizziness, flushing

Occurring in < 0.5% of the Luminity-dosed patients:

Body as a Whole:	fatigue, fever, hot flushes, pain, rigors and syncope
Cardiovascular:	abnormal ECGs, bradycardia, tachycardia, palpitation, hypertension and hypotension
Digestive System:	dyspepsia, dry mouth, tongue disorder, toothache, abdominal pain, diarrhoea and vomiting
Hematology:	granulocytosis, leukocytosis, leukopenia, monocytosis eosinophilia
Musculoskeletal:	arthralgia
Nervous System:	leg cramps, hypertonia, vertigo and paraesthesia
Haemic and Lymphatic:	haematoma, lymphadenopathy
Respiratory System:	coughing, hypoxia, pharyngitis, rhinitis and dyspnoea.
Special Senses:	decreased hearing, conjunctivitis, abnormal vision and taste perversion
Skin:	pruritus, rash, erythematous rash, urticaria, increased sweating, dry skin
Urinary:	albuminuria and abnormal urine
Laboratory Abnormalities:	increased bilirubin, AST/SGOT, SGPT/ALT, creatine phosphokinase, LDH, creatinine, glucose and non-protein nitrogen

Adverse Events in Echocardiographic Studies

All New-Onset AEs Occurring in at least 0.5% of Luminity-treated Subjects in All Echocardiography Studies

Preferred Term	All AEs		Treatment-Related AEs	
Total Subjects Treated	1558		1558	
Total Subjects with AEs	475	(30.5%)	106	(6.8%)
Fatigue	144	(9.2%)	3	(0.2%)
Dyspnoea	75	(4.8%)	0	
Headache	60	(3.9%)	34	(2.2%)
Chest pain	54	(3.5%)	3	(0.2%)
Flushing	25	(1.6%)	11	(0.7%)
Nausea	24	(1.5%)	15	(1.0%)
Dizziness	17	(1.1%)	8	(0.5%)
Chest discomfort	16	(1.0%)	0	
Back pain	12	(0.8%)	8	(0.5%)
Hypertension NOS	11	(0.7%)	0	
Arthralgia	9	(0.6%)	0	
Electrocardiogram abnormal NOS	9	(0.6%)	2	(0.1%)
Hyperglycaemia NOS	9	(0.6%)	3	(0.2%)
Diarrhoea NOS	8	(0.5%)	3	(0.2%)
Injection site reaction NOS	8	(0.5%)	2	(0.1%)
Pain NOS	8	(0.5%)	0	

The most frequent new-onset AEs (≥1%) in Luminity-treated echocardiography subjects were fatigue, dyspnoea, headache, chest pain or discomfort, flushing, nausea and dizziness. The new-onset AEs of fatigue, dyspnoea and chest pain were observed mainly in the studies using exercise stress testing

In the two most frequently used bolus dosage categories (10-20 µl/kg and >20-40 µl/kg), the overall new-onset AE frequency was similar (27.0% vs 27.3%) and was comparable to that for subjects in echocardiography studies as a whole (30.5%). Only one of ten echocardiography subjects who received a Luminity bolus dosage greater than 40µl/kg experienced any new-onset AEs.
No dose relationship could be detected for the overall frequency of treatment-related new-onset AEs in echocardiography subjects receiving bolus doses of Luminity. The overall frequency of treatment-related new-onset AEs in the 10-20 µl/kg bolus dosage category was 13.0%. The only treatment-related new-onset AEs that occurred in ≥1% of echocardiography subjects of this dosage category were headache (5.2%), nausea (2.1%), dizziness (1.6%), flushing (1.3%), vomiting NOS (1.0%), and back pain (1.0%).

Chest pain was reported by a total of 37 Luminity -treated subjects receiving various dosages of Luminity as an infusion in echocardiography studies. The AE was rated as mild or moderate in 35 of the 37 cases. No relationship was observed between dose of Luminity and occurrence of chest pain, nor between dose of Luminity and intensity of chest pain. In 34 of the 37 subjects, the chest pain occurred shortly after the start of infusion. However, all the cases of chest pain were rated as unlikely to be related to use of Luminity.

- Serious adverse event/deaths/other significant events

Number (%) of Luminity-treated Subjects With Serious New-Onset Adverse Events in Echocardiography and Radiology Studies

Category	Echocardiography		Radiology		Combined	
Total Subjects Treated	1558		968		2526	
Total Subjects with AEs	475	(30.5%)	171	(17.7%)	646	(25.6%)
Total Subjects with Serious AEs	19	(1.2%)	14	(1.4%)	33	(1.3%)
Subjects with Serious AEs Leading to Death	3	(0.2%)	5	(0.5%)	8	(0.3%)
Subjects with Non-fatal Serious AEs	16	(1.0%)	9	(0.9%)	25	(1.0%)

Eight (0.3%) of the 2526 subjects treated with Luminity in echocardiography and radiology studies died. All of the deaths occurred several days after administration of Luminity and were considered by the investigators as unlikely to be related to Luminity dosing. Most of the subjects were elderly, and had serious cardiac illnesses and/or cancer. One subject was a 33-year old black man with a history of CHF, idiopathic dilated cardiomyopathy and heart transplant.

Overall, 33 (1.3%) of the 2526 Luminity -treated subjects in echocardiography and radiology studies had serious AEs. None of the serious AEs was classed as treatment-related.

Exercise Stress Studies

In the exercise stress studies, nearly all new-onset AEs were classed as stress-related. The overall rate of stress-related AEs was higher in control subjects.

Dobutamine Stress Study DMP 115-024

The only new-onset AEs that occurred in more than one subject of a category and treatment were nausea, vomiting NOS and chest pain. The only new-onset AEs classed as not stress-related in Luminity-treated subjects were chest pain (2 subjects), feeling jittery (1 subject), gait abnormal (1 subject), liver function test abnormal (1 subject) and wheezing

Cardiac

None of the serious cardiac AEs in Luminity-treated subjects were considered to be attributable to the administration of Luminity.

- Laboratory findings

Coagulation was not assessed in the pivotal studies, since no clinically meaningful changes were observed in early clinical trials. Subjects had routine urinalysis which revealed no clinically significant changes in urinalysis in Luminity-treated subjects vs placebo-treated subjects.

Haematology and Serum Chemistry

In pivotal trials in general, the mean changes observed in haematology and serum chemistry parameters from baseline to 24 hours were similar among subjects who received either placebo, 10 to 20 µl/kg Luminity, or >20 µl/kg Luminity.

Vital Signs

Overall, in the combined group of all studies, about two-thirds to three quarters of all Luminity-treated subjects had changes of ≤20% in BP, pulse and respiration rate from baseline

Review of the individual BP data for the subjects with hypertension or hypotension as an AE indicated that the findings reported as AEs were transient and BP values returned to normal in all cases.

Electrocardiogram (ECG)
None of the subjects with an absolute increase in QTc interval ≥30 msec from baseline experienced any rhythm disorders in association with the increase in QTc. No cases of QTc prolongation were reported as a new-onset AE.

ECG parameters for doses up to 10 ml/kg were monitored in subjects at multiple time points from 1 hour to 72 hours after the first bolus injection. QTc prolongation associated cardiac rhythm changes were not considered to be clinically important.

- Safety in special populations

Populations evaluated were demographic groups and those with medical histories of CHF or COPD.

Gender
The overall incidence of new-onset AEs was similar for both gender groups of Luminity-treated subjects in the echocardiography studies.
The overall incidence of new-onset AEs was higher in Luminity-treated women than in Luminity-treated men in the radiology studies.

Age
The overall incidence of new-onset AEs was similar for both age groups (<65, >65 years old) of Luminity-treated subjects in echocardiography as well as in radiology studies.

Chronic Obstructive Pulmonary Disease
Treatment-related new-onset AEs occurred at similar rates in both Luminity-treated COPD subjects and Luminity-treated subjects without COPD. In Luminity-treated COPD subjects, the only treatment-related new-onset AEs were one case each of palpitations, dysgeusia and flushing.

Congestive Heart Failure
In the placebo controlled studies the overall frequency of AEs appears to be higher in Luminity-treated CHF subjects than in placebo CHF subjects. However, no difference was seen between the treatments with respect to the overall rate of treatment-related new-onset AEs in CHF subjects.

- Safety related to drug-drug interactions and other interactions

Drug-drug interaction studies have not been performed with Luminity.

- Discontinuation due to adverse events

The 19 Luminity-treated subjects who did not complete the study due to AEs comprised 11 (0.7%) of 1558 Luminity-treated subjects in echocardiography studies and 8 (0.8%) of 968 Luminity-treated subjects in radiology studies.

Most of the AEs resulting in study non-completion were transient, resolving within 1 minute to 2.5 hours; none were serious; almost all were considered possibly or probably related to use of Luminity.

- Post marketing experience

Data available for the time period from 1 October 2001 to 30 July 2004 indicated that, the approximate total number of vials of DMP 115 distributed both commercially and through sales were 338,883.

Review of the safety data from marketed use of Luminity presented in the period from 28 December 2000 until 13 September 2004, inclusive, did not reveal any important new safety issues or trends associated with Luminity therapy since the product was first marketed

- Discussion on clinical safety

Approximately 25% of all patients who received Luminity experienced adverse events (fatigue, headache, dyspnoea and chest pain). The incidence of adverse effects was both dose related and increased by faster rates of administration. Adverse events occurred more frequently in those having infusions. Chest pain, although not considered to be drug related, did at least have a temporal association to administration.

Eight (0.3%) of the 2526 subjects treated with Luminity died. All of the deaths occurred several days after administration of Luminity and were considered as unlikely to be related to Luminity dosing. Most of the subjects were elderly, and had serious cardiac illnesses and/or cancer. One subject was a 33-year old black man with a history of CHF, idiopathic dilated cardiomyopathy and heart transplant. 2 additional cases were reported during post marketing.

Overall, 33 (1.3%) of the 2526 Luminity-treated had serious AEs. None of them was classed as treatment-related. Chest pain and ECG abnormality occurred more frequently than in comparative groups. However, the patient population might lead to an overall higher incidence of cardiovascular effects.

Important identified risks are the administration of Luminity to patients with impaired pulmonary function.

Important missing information is data of Luminity administration to a sufficient number of patients with COPD or CHF.
In different studies, the mechanical index values varied. It is known that high ultrasound mechanical index values may cause microsphere cavitation or rupture and lead to ventricular arrhythmias. Additionally, end-systolic triggering with high mechanical indices has been reported to cause ventricular arrhythmias. The safety of activated Luminity at high mechanical indices and with the use of end-systolic triggering has not been established.

There is no experience of acute over-dosage with Luminity in humans. There is no specific antidote for over-dosage with Luminty and treatment of overdose should consist of general supportive measures.

5. Pharmacovigilance

Detailed description of the Pharmacovigilance system

The CHMP considered that the Pharmacovigilance system as described by the applicant fulfils the legislative requirements.

Risk Management Plan

The MAA submitted a risk management plan, which included a risk minimisation plan.

Table Summary of the risk management plan

Safety issue	Proposed pharmacovigilance activities	Proposed risk minimisation activities
Identified risks of allergic and anaphylactic reactions	• Actively follow-up all potentially severe allergic or anaphylactic reactions that occur during Luminity administration by using a specialised questionnaire to obtain more complete and consistent information for each report of potentially severe allergic or anaphylactic reactions, to make a more informed causality assessment and to obtain sufficient information for stratification purposes • Monitor all the new reports of anaphylactic or allergic reactions. Any significant changes in frequency will be described in the PSUR with appropriate modifications to product labeling • Monitor the follow-up process in order to determine if there is need for improvement • The review of potentially severe allergic or anaphylactic cases will be presented with the submission of each PSUR, annual reassessment, and license renewal • If at any time during the marketing of Luminity, the Company recognises a significant issue with severe allergic or anaphylactic reactions in patients receiving Luminity, a complete assessment of these reports will be conducted	The risks of Luminity are not of a particular nature or seriousness that would require specific risk minimisation measures beyond careful use of labeling and packaging including suitable warnings in the product literature • Luminity should only be administered by trained physicians with technical expertise in performing and interpreting contrast echocardiograms as per Section 4.2 of the SPC • Contraindication for patients with known hypersensitivity to the active substance or to any of the excipients in Section 4.3 of the SPC Listed as ADR in Section 4.8 of the SPC
Potential risk of fatal outcomes	• Actively follow-up all fatal cases that occur during Luminity administration by using a specialised questionnaire to obtain more complete and consistent information for each report with a fatal outcome and to gather sufficient information	The risks of Luminity are not of a particular nature or seriousness that would require specific risk minimisation measures beyond careful use of labeling and packaging including suitable warnings in the product literature • Luminity should only be administered by trained physicians with technical expertise in performing and

	to make a more informed causality assessment • Monitor all the new reports of fatal outcomes. Any significant changes in frequency will be described in the PSUR with appropriate modifications to product labeling. • Monitor the follow-up process in order to determine if there is need for improvement • The review of fatal outcomes will be presented with the submission of each PSUR, annual reassessment, and license renewal • If at any time during the marketing of Luminity, the Company recognises a significant issue with fatal outcomes in patients receiving Luminity, a complete assessment of these reports will be conducted	interpreting contrast echocardiograms as per Section 4.2 of the SPC • Contraindication for patients with known hypersensitivity to the active substance or to any of the excipients in Section 4.3 of the SPC Warnings in Section 4.4 of the SPC

The CHMP, having considered the data submitted in the application, is of the opinion that no additional risk minimisation activities are required beyond those included in the product information.

6. Overall conclusions, risk/benefit assessment and recommendation

Quality

The quality of this product is considered to be acceptable when used in accordance with the conditions defined in the SPC. Physicochemical and biological aspects relevant to the uniform clinical performance of the product have been investigated and are controlled in a satisfactory way.

Non-clinical pharmacology and toxicology

Overall, the primary pharmacodynamic studies provided adequate evidence, that Luminity is an effective contrast agent to use in conjunction with ultrasound in order to aid in the diagnosis and assessment of myocardial perfusion defects. A correlation demonstrated increased acoustic power and increased pressure shortens the persistence of microbubbles in circulation.
No secondary pharmacodynamic studies were performed.

From the pharmacokinetic point of view, the beagle dog was the most relevant species for non-clinical efficacy and safety studies because of the established use of dogs in pulmonary expiration studies. PFP expired through the lungs is unchanged. There is no indication of any long-term retention or potential for accumulation of the non-endogenous components. The rapid elimination of PFP in the expired air is consistent with the rapid disappearance of ultrasound contrast after activated Luminity administration.

Overall, the preclinical toxicology programme revealed no special hazard for humans based on conventional studies of genotoxicity, fertility, embryo/foetal development, parturition or post-natal development, and local tolerance.

The lack of carcinogenicity studies is considered justified, since genotoxicity studies did not indicate potential of mutagenicity or clastogenicity [5].

Efficacy

The demonstration of efficacy of Luminity for the use as an ultrasound contrast-enhancing agent in echocardiography is based on 5 pivotal and 7 supportive studies.

The five pivotal studies support the use of Luminity for contrast-enhanced echocardiographic imaging of cardiac structure (ventricular chambers and endocardial borders) using fundamental imaging by demonstrating a significant benefit of contrast-enhanced ultrasound to unenhanced ultrasound.

Data from two Phase 2 studies (DMP 115-018 and 022) support the use of Luminity in imaging ventricular chambers and endocardial border delineation during stress/rest echocardiography using non-linear imaging in subjects with known or suspected coronary artery disease (CAD).

In addition data presented during the assessment from two further studies (DMP 115-401 and -410) support the claimed indication using non-linear ultrasound technique. DMP 115-410 shows, that the overall proportions of visualised cardiac segments were significantly higher for Luminity-enhanced echocardiography than for unenhanced echocardiography. DMP 115-401 demonstrates, that significantly more images were diagnostic following enhancement, confidence was significantly greater with enhancement and that less additional diagnostic testing was requested following contrast enhanced imaging.

Safety

The incidence of adverse effects (25% of all treated patients) was both dose related and increased by faster rates of administration. Adverse events occurred more frequently in those having infusions.

All of the deaths 8/2526 (0.3%) occurred several days after administration of Luminity and were considered as unlikely to be related to Luminity dosing. Most of the subjects were elderly, and had serious cardiac illnesses and/or cancer.

33 of the 2526 Luminity-treated patients (1.3%) had serious AEs. None of them was classed as treatment-related. Chest pain and ECG abnormality occurred more frequently than in comparative groups.

Important identified risks are the administration of Luminity to patients with impaired pulmonary function.

Important missing information is data of Luminity administration to patients with COPD or CCF.

From the safety database all the adverse reactions reported in clinical trials and post-marketing have been included in the Summary of Product Characteristics.

Having considered the safety concerns in the risk management plan, the CHMP considered that the proposed activities described in section 3.5 adequately addressed these.

The applicant provided an acceptable justification for not performing a user consultation testing of the package leaflet.

Benefit-risk assessment

Overall the administration of Luminity is safe. For the echocardiography Indication, the lack of comparator studies can be dealt with by considering the available indirect comparisons, and the applicant undertaking a post approval study commitment to perform a study to make a direct comparison between Luminity and SonoVue.
Despite the fact that studies DMP 115-018 and DMP 115-022 failed primary efficacy analysis based on the comparison of the ability of at least two blinded readers to detect exercise stress-induced wall motion abnormalities from echocardiography using harmonic imaging techniques and classification of CAD based on angiographic findings, the study was successful in an analysis of wall segment evaluability, number of evaluable segments per subject and by per segment analysis, which was not planned in the protocol.
The results of additional studies such as DMP 115-410 using harmonic imaging offer supportive information that the overall proportions of visualised cardiac segments were significantly higher for Luminity-enhanced echocardiography than for unenhanced echocardiography and the results of DMP 115-410 seem to support an increase in the proportion of visualised segments following contrast administration.
The results of DMP 115-401 suggest that significantly more images were diagnostic following enhancement, confidence was significantly greater with enhancement and that less additional diagnostic testing was requested following contrast enhanced imaging.

Taking the safe use and the evidence of efficacy submitted into account, the risk-benefit of Luminity in the claimed echocardiography indication is positive.

A risk management plan was submitted. The CHMP, having considered the data submitted, was of the opinion that routine pharmacovigilance was adequate to monitor the safety of the product.

Recommendation

Based on the CHMP review of data on quality, safety and efficacy, the CHMP considered by consensus that the benefit-risk balance of Luminity in the following approved indication;

> This medicinal product is for diagnostic use only. Luminity is an ultrasound contrast-enhancing agent for use in patients in whom non-contrast echocardiography was suboptimal (suboptimal is considered to indicate that at least two of six segments in the 4- or 2-chamber view of the ventricular border were not evaluable) and who have suspected or established coronary artery disease, to provide opacification of cardiac chambers and improvement of left ventricular endocardial border delineation at both rest and stress.

was favourable and therefore recommended the granting of the marketing authorisation.

References

1. Metzger-Rose C, Krupinski EA, Wright WH, Baker MR, McCreery TP, Barrette TR, Unger EC: **Ultrasonographic detection of testicular ischemia in a canine model using phospholipid coated microbubbles (MRX-115)**. *J Ultrasound Med* 1997, **16**(5):317-324; quiz 325-316.
2. Maruyama H, Matsutani S, Sato G, Nakano Y, Mitsuhashi O, Yoshikawa M, Ebara M, Saisho H, Ohto M: **Enhanced color flow images in small hepatocellular carcinoma**. *Abdom Imaging* 2000, **25**(2):164-171.
3. Boyden PA, Dresdner KP, Jr.: **Electrogenic Na(+)-K+ pump in Purkinje myocytes isolated from control noninfarcted and infarcted hearts**. *Am J Physiol* 1990, **258**(3 Pt 2):H766-772.
4. Hutter JC, Luu HM, Mehlhaff PM, Killam AL, Dittrich HC: **Physiologically based pharmacokinetic model for fluorocarbon elimination after the administration of an octafluoropropane-albumin microsphere sonographic contrast agent**. *J Ultrasound Med* 1999, **18**(1):1-11.
5. EMEA: **ICH Topic S1B Carcinogenicity: Testing for Carcinogenicity of Pharmaceuticals**. 1998:http://www.emea.eu.int/pdfs/human/ich/029995en.pdf.