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RESEARCH ARTICLE

β3 adrenergic agonist treatment in chronic pulmonary hypertension associated with heart failure (SPHERE-HF): a double blind, placebo-controlled, randomized clinical trial

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Aims

Pulmonary hypertension (PH) associated with left heart disease is an increasingly prevalent problem, orphan of targeted therapies, and related to a poor prognosis, particularly when pre- and post-capillary PH combine. The current study aimed to determine whether treatment with the selective β3 adrenoreceptor agonist mirabegron improves outcomes in patients with combined pre- and post-capillary PH (CpcPH).

Methods and results

The β3 Adrenergic Agonist Treatment in Chronic Pulmonary Hypertension Secondary to Heart Failure (SPHERE-HF) trial is a multicentre, randomized, parallel, placebo-controlled clinical trial that enrolled stable patients with CpcPH associated with symptomatic heart failure. A total of 80 patients were assigned to receive mirabegron (50 mg daily, titrated till 200 mg daily, $n = 39$) or placebo ($n = 41$) for 16 weeks. Of them, 66 patients successfully completed the study protocol and were valid for the main analysis. The primary endpoint was the change in pulmonary vascular resistance (PVR) on right heart catheterization. Secondary outcomes included the change in right ventricular (RV) ejection fraction by cardiac magnetic resonance or computed tomography, other haemodynamic variables, functional class, and quality of life. The trial was negative for the primary outcome (placebo-corrected mean difference of 0.62 Wood units, 95% confidence interval [CI] −0.38, 1.61, $p = 0.218$). Patients receiving mirabegron presented a

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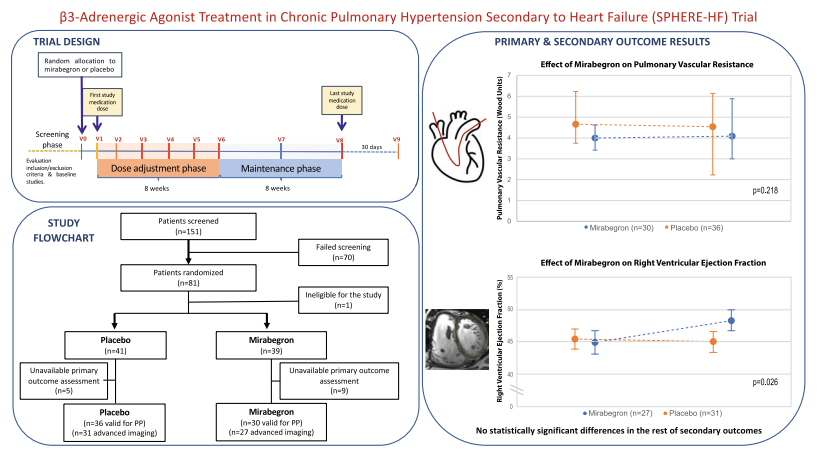
significant improvement in RV ejection fraction as compared to placebo (placebo-corrected mean difference of 3.0%, 95% CI 0.4, 5.7%, $p = 0.026$), without significant differences in other pre-specified secondary outcomes.

Conclusions SPHERE-HF is the first clinical trial to assess the potential benefit of β3 adrenergic agonists in PH. The trial was negative since mirabegron did not reduce PVR, the primary endpoint, in patients with CpcPH. On pre-specified secondary outcomes, a significant improvement in RV ejection fraction assessed by advanced cardiac imaging was found, without differences in functional class or quality of life.

Graphical Abstract



Summary of the study outline, flowchart, and results of the primary and secondary outcomes. The β3 selective agonist mirabegron did not improve pulmonary haemodynamics in adult patients with combined pre- and post-capillary pulmonary hypertension associated with symptomatic heart failure. On secondary outcomes, a significant improvement in right ventricular ejection fraction assessed by advanced cardiac imaging was found, without differences in functional class or quality of life. PP, per protocol population.

Keywords Pulmonary hypertension • Heart failure • Mirabegron • β3 adrenoreceptors

Introduction

Pulmonary hypertension (PH) commonly complicates heart failure (HF) and confers a bad prognosis.[1,2] Up to two thirds of patients with HF with both reduced (HFrEF) or preserved ejection fraction (HFpEF) develop PH.[3–5] Of them, approximately 13% progress to combined pre- and post-capillary PH (CpcPH), characterized by higher pulmonary vascular resistance (PVR), more severe pulmonary vascular remodelling and worse prognosis than isolated post-capillary PH.[3] Right ventricular (RV) function is a well-recognized prognostic factor in PH associated with left heart disease (LHD), with an impact even greater than the severity of the increase in PVR.[1] Currently there are not specific pharmacological therapies approved for patients with CpcPH either to improve pulmonary haemodynamics or RV adaptation.[6] Beta-3 adrenoreceptors (β3AR) are expressed in the human myocardium[7] and vessels[8] and have been described to be upregulated in LHD.[7] Their stimulation or overexpression has been shown to cause vasodilatation[9,10] and prevention of cardiac remodelling in experimental animal models.[11–13] In addition, prior experimental research has demonstrated the presence of mRNA expression of hB3AR in human pulmonary arteries and has shown that treatment with β3AR agonists produced a beneficial effect on pulmonary haemodynamics and RV performance, associated with an attenuation in pulmonary vascular proliferation in a swine model of chronic post-capillary PH.[14] The selective oral β3AR agonist mirabegron is currently approved for the treatment of overactive bladder syndrome and has demonstrated a good safety profile in healthy subjects and patients, both suffering from the urinary condition[15–17] or HFrEF.[18] The potential efficacy of treatment with β3AR agonists on patients affected by PH has never been evaluated. On the basis of these considerations, the β3 Adrenergic Agonist Treatment in Chronic Pulmonary Hypertension Secondary to Heart Failure (SPHERE-HF) trial was set up to evaluate the efficacy and safety of the selective β3AR agonist mirabegron in patients with CpcPH.

Our hypothesis was that treatment with mirabegron in patients with CpcPH would result in a beneficial effect due to: (i) a reduction in PVR, (ii) an increase in RV performance, and (iii) improvement in clinical status without an increase in severe adverse events (AEs).

Methods

Study design

The SPHERE-HF trial was a Phase II double-blind multicentre, with balanced randomization (1:1), placebo-controlled, parallel-group clinical trial conducted in Spain (five sites). The study received ethical approval from regional and national health service research ethics committees and was conducted in accordance with the principles of good clinical practice. All patients provided written informed consent before randomization. The Hospital Clínic de Barcelona coordinated the trial in collaboration with the Centro Nacional de Investigaciones Cardiovasculares (CNIC). An independent data safety monitoring board reviewed safety data every 6 months throughout the trial. This trial was registered at ClinicalTrials.gov NCT02775539 and EudraCT: 2016-002949-32. Details of the trial design have been previously reported.[19]

Participants

Patients with PH associated with HF were screened for possible inclusion. Eligible participants were adults aged 18 or over with symptomatic HF (New York Heart Association [NYHA] functional class II–III) and secondary CpcPH who were on optimized evidence-based pharmacological treatment and stable clinical condition during the 30 days preceding recruitment. Inclusion and exclusion criteria are presented on *Table 1*. The exclusion criteria of QTc interval on electrocardiogram (ECG) >430 ms in men and >450 ms in women, that had been established as a general safety standard, was modified by an amendment to QTc >480 ms, based on the high percentage of patients with CpcPH who presented this exclusion criteria at baseline and the absence of data suggesting that mirabegron at the doses used could significantly increase QTc.

Manufacturing of study medication

Manufacturing of mirabegron and placebo was carried out at the Hospital Clínic Pharmacy department. The excipient for placebo capsules (mixture of microcrystalline cellulose and colloidal silica) was received from the manufacturer (Fagron Iberica S.A.U, Terrassa, Spain) and included in capsules. Similarly, mirabegron pills were bought from Astellas Pharma (Chuo, Tokyo, Japan), taken out of the blisters, and automatically encapsulated (a single pill into a capsule). Immediately, capsules containing mirabegron or excipient (exactly the same appearance and taste) were introduced in polyethylene bottles (30 capsules each), labelled as study medication with the batch number and expiration, and kept in the clinical trials area in conservation conditions (atmosphere temperature <25°C). Labelled was done in a blinded manner to guarantee masking of patients and physicians. Extra tablets were given at each clinical visit. Therapeutic compliance was evaluated by counting empty blisters and remaining tablets.

Randomization and blinding

Patients were randomly allocated (1:1) to mirabegron or placebo using randomly selected block sizes stratified by center using the Blockrand package (R Foundation). Generation of the randomization list and allocation concealment were done by statisticians who were independent from researchers and the people involved in the implementation of assignments. The randomization list was provided exclusively to the pharmacy department, which was responsible for the preparation of medication kits by identifying them with a unique sequential number for the entire study and provide them to the centres. Researchers assigned medication kits in a sequential order and recorded the kit number provided to each patient in the data collection system.

Procedures

Patients underwent the following baseline procedures and assessments within 4 weeks before random allocation: demographic and medical history data collection; physical examination (including blood pressure, heart rate, and pulse oxymetry); NYHA functional class; blood sample analysis including N-terminal pro-B-type natriuretic peptide (NT-proBNP); ECG; echocardiography; right heart catheterization (RHC); 6-min walking test, and a cardiac magnetic resonance (CMR) or a cardiac computed tomography (CCT). CMR was used preferably but in patients with severe claustrophobia or cardiac devices, a dedicated CCT examination was performed instead to measure RV volumes and function. The protocol for the RHC was previously written and standardized in all participating hospitals. Briefly, the procedure was performed in a supine position with patients breathing mostly room air or supplementary oxygen when needed through a venturi mask with a fixed FiO_2. Zero reference level was established at mid thoracic level, at the intersection of the frontal plane at the mid thoracic level, and the transverse plane at the level of fourth anterior intercostal space. Determination of all pressure values was an averaged over 3–5 respiratory cycles (continuous registry). Systemic blood pressure was firstly measured, followed by the measurement of systolic, diastolic and mean pulmonary artery pressure (PAP), pulmonary artery wedge pressure (PAWP, including 'v' wave when existed) and right atrial pressure. When patients were in atrial fibrillation a longer stretch of the record was analysed. Determination of cardiac output was made by thermodilution at least three times without a difference greater than 10%. Finally, a sample from the pulmonary artery was drawn to measure PvO_2 and SvO_2. A complete description of the examinations, haemodynamic and imaging protocols have been previously published.[19]

Eligible patients were randomized to receive mirabegron (50 mg) or placebo once daily on Visit 0, started study medication on Visit 1 (during the 5 days after randomization), and were reviewed 1 week later in a safety visit (Visit 2), all in a blinded fashion. Thereafter, medication dose was uptitrated every 2 weeks for 8 weeks (up to a maximum of 200 mg daily) based on patients' blood pressure, heart rate, QTc interval, blood analysis and clinical status assessed at each visit, and then maintained for another 8 weeks. Once finished the 16-week period under treatment, patients underwent the same study examinations performed at baseline and stopped the study medication. A final security monitoring visit took place 30 days after the last study medication dose, following clinical trials regulation. *Figure 1* shows the scheme of the study conduct and procedures.

Outcomes

The primary outcome was the change from baseline to week 16 in PVR on RHC, calculated in Wood units (WU), as the difference

Table 1 Inclusion and exclusion criteria

Inclusion criteria	Exclusion criteria
Written informed consent	Non-coronary cardiac surgery (e.g. valvular surgery) or non-coronary structural percutaneous procedure (e.g. percutaneous mitral repair) within the 12 months preceding recruitment or scheduled
≥18 years old	Myocardial infarction or coronary revascularization within the 3 months preceding recruitment
HF with reduced, mildly reduced or preserved LVEF, according to the definition of the European Society of Cardiology guidelines.[6]	CRT implantation within the 6 months preceding recruitment

Combined pre- and post-capillary PH determined by RHC showing the following:
• PAWP or LVEDP ≥15 mmHg.
• Mean PAP ≥25 mmHg; and:
  • PVR ≥3 WU and/or diastolic gradient ≥7 mmHg, or
  • Transpulmonary gradient ≥12 mmHg
	Sinus tachycardia or uncontrolled atrial fibrillation (HR >100 bpm)

NYHA functional class II–IV	Uncontrolled systemic hypertension (systolic BP >180 mmHg or diastolic BP >110 mmHg) or symptomatic hypotension (systolic BP <90 mmHg)
On optimized evidence-based pharmacological treatment.	Diagnosis of infiltrative cardiomyopathy
Stable clinical condition defined as no changes in therapeutic regimen for HF or hospitalization in the 30 days preceding recruitment and no current plan for changing therapy	Pre-menopausal women who have not undergone total hysterectomy
	Expected survival <1 year due to a disease other than HF
	Severe renal failure (GFR < 30 ml/min/1.73 m^2)
	Severe hepatic impairment (transaminase elevation >3 times ULN)
	Prolonged cQT interval on the ECG (>430 ms in male or >450 ms in female)[a]
	Concomitant use with specific pulmonary vasodilators (sildenafil, bosentan, macitentan, riociguat or other endothelin receptor blockers, phosphodiesterase 5 inhibitors or guanylate cyclase stimulators)
	Treatment with digoxin, flecainide, propafenone, dabigatran, tricyclic antidepressants or other CYP2D6 inhibitors (other than beta-blockers)
	Severe COPD (FEV_1/FVC ratio <0.7 together with FEV_1 <50% predicted value)
	Severe restrictive lung disease (TLC <50%)
	Participation in another clinical trial
	Known allergy to mirabegron or any of the excipients

BP, blood pressure; COPD, chronic obstructive pulmonary disease; CRT, cardiac resynchronization therapy; FEV_1, forced expiratory volume in the first second; FVC, forced vital capacity; GFR, glomerular filtration rate; HR, heart rate; LVEDP, left ventricular end-diastolic pressure; NYHA, New York Heart Association; PAP, pulmonary artery pressure; PAWP, pulmonary artery wedge pressure; PVR, pulmonary vascular resistance; RHC, right heart catheterization; TLC, total lung capacity; ULN, upper limit of normal; WU, Wood units.

[a]This criterion was changed by an amendment to QTc >480 ms.

between mean PAP (mmHg) and PAWP (mmHg), divided by the cardiac output (l/min). Pre-specified secondary outcomes were the change from baseline in: RV ejection fraction assessed by advanced imaging (CMR or CCT), NYHA functional class, 6-min walk distance, dyspnoea Borg scale score, quality of life evaluated by the Kansas City Cardiomyopathy Questionnaire overall summary score, mean PAP, transpulmonary gradient, diastolic gradient, cardiac output, and NT-proBNP. Safety measures included HF decompensation, death, urgent heart transplantation, AEs and adverse drug reactions, as well as monitoring of heart rate and the QTc interval on ECG. An independent data safety monitoring board reviewed all major events and determined the potential relationship with study medication, according to standard operational procedures. An imaging core-lab centered in CNIC measured all acquired images (echocardiograms, CMR and CCT) in a completely blinded fashion and reported directly to the external statistics board.

Statistical analysis

As described previously, assuming a standard deviation of 3.2 WU (based in a population with CpcPH)[20] and a correlation between baseline and 16-week measurement of 0.6, a sample size of 31 subjects per group was required to detect a placebo-adjusted difference of 2.1 WU, with a power of 80% and a 2-sided significance level of 5%. Anticipating a 20% dropout rate, the estimated number of patients needed was 80. Calculations were made using the

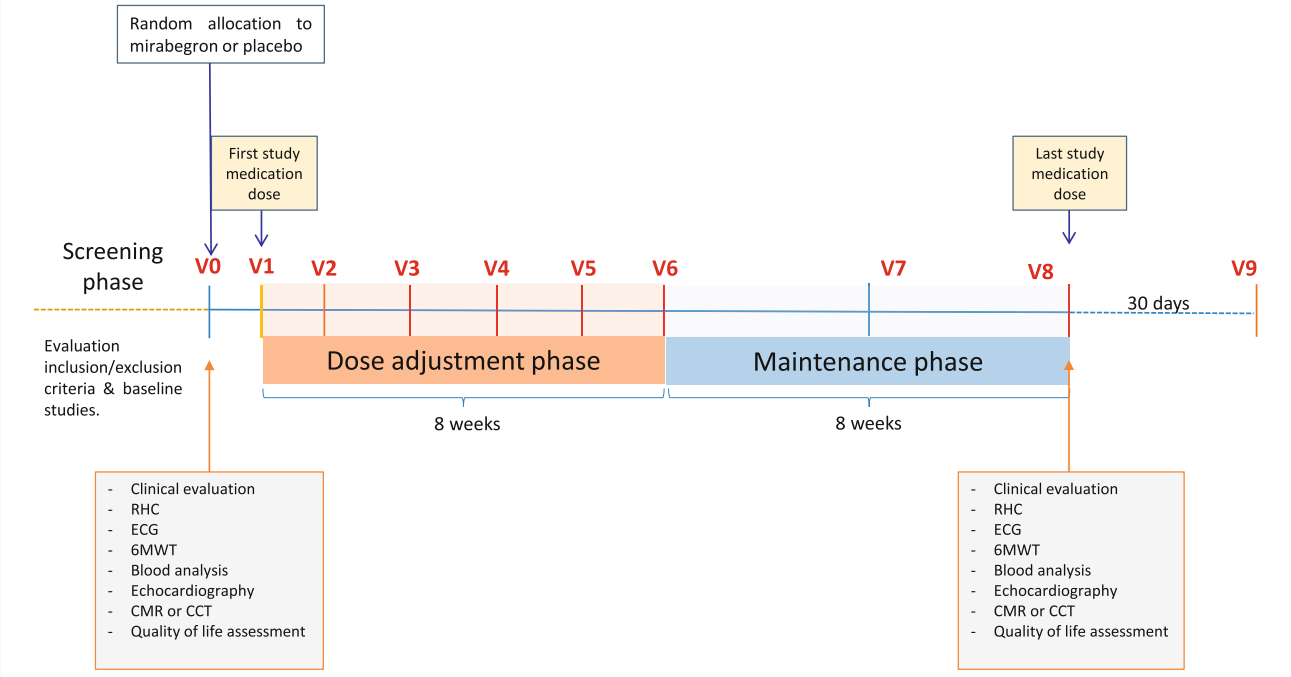


Figure 1 Study conduct and procedures. Summary of the study conduct and procedures performed at baseline and after 16 weeks of study medication (mirabegron or placebo). 6MWT, 6-min walking test; CCT, cardiac computed tomography; CMR, cardiac magnetic resonance; ECG, electrocardiogram; RHC, right heart catheterization.

power.t.test function available in R software (R Foundation, Vienna, Austria).

The following analysis populations were predefined: intention to treat (ITT) population (all randomized patients); per protocol (PP) population (patients from the ITT who have the primary outcome measured at 16 weeks and who took at least 80% of all medication doses); and the safety population (all patients who took at least one dose of the assigned treatment).

As pre-established in the statistical analysis plan (and previously published[19]), due to the nature of the trial (proof-of-concept) and the selected primary event (PVR on RHC), the primary analysis was performed in the PP population, with supportive analysis in patients on the ITT population. For the ITT population, missing values in the final measure of the primary outcome were replaced by the baseline value (last observation carried forward). The treatment effect on the primary and pre-specified secondary outcomes was analysed using an ANCOVA model that included the baseline value, treatment, and baseline × treatment interaction. Interaction term was removed if the Wald test was not statistically significant ($p > 0.10$). The centre was not included in the model due to small centre sizes.[21] Model conditions were investigated in all cases (normality of residuals by Shapiro–Wilk test and the presence of observations with influence by graphical methods). In case of deviations from normality or observations with influence, the change from baseline to final measure (delta) was analysed by randomization test and bootstrapping (9999 replicates), as implemented in the boot.t.test function of the R MKinfer package. In case of NYHA functional class, logistic regression instead of ANCOVA was used. In addition, patients were analysed by protocol-pre-specified subgroups according to left ventricular ejection fraction (<40% vs. ≥40%) and the maximum tolerated dose (mg/dl). Safety analyses were performed descriptively in the safety population. Data are presented as mean ± standard deviation or median (first–third quartile) as corresponding. A p-value < 0.05 was considered as statistically significant. All statistical analysis were undertaken using R package (Vienna, Austria).

Results

Patients

Of the 151 patients screened from June 2017 to December 2020, 81 were initially considered eligible for randomization (*Figure 2*). One patient was excluded after randomization because a lung ventilation–perfusion scan revealed a segmental perfusion defect raising doubts about the potential thromboembolic origin of PH. Of the 80 patients who were included in the ITT population, 67 underwent a second RHC at 16 weeks and all of them but one achieved at least 80% of therapeutic compliance, so these 66 patients composed the PP population (30 allocated to mirabegron and 36 to placebo). Reasons for withdrawal are summarized in *Figure 2*.

Demographic and clinical characteristics were well balanced between treatment groups at baseline (*Table 2*). Overall, nearly two thirds of the patients had HFpEF and non-ischaemic aetiology. About half of the patients had marked limitation of physical activity (NYHA functional class III). Approximately one third ($n = 19$) had previous non-coronary cardiac surgery (aortic and/or mitral valve surgery in 18 patients, and septal myectomy in one)

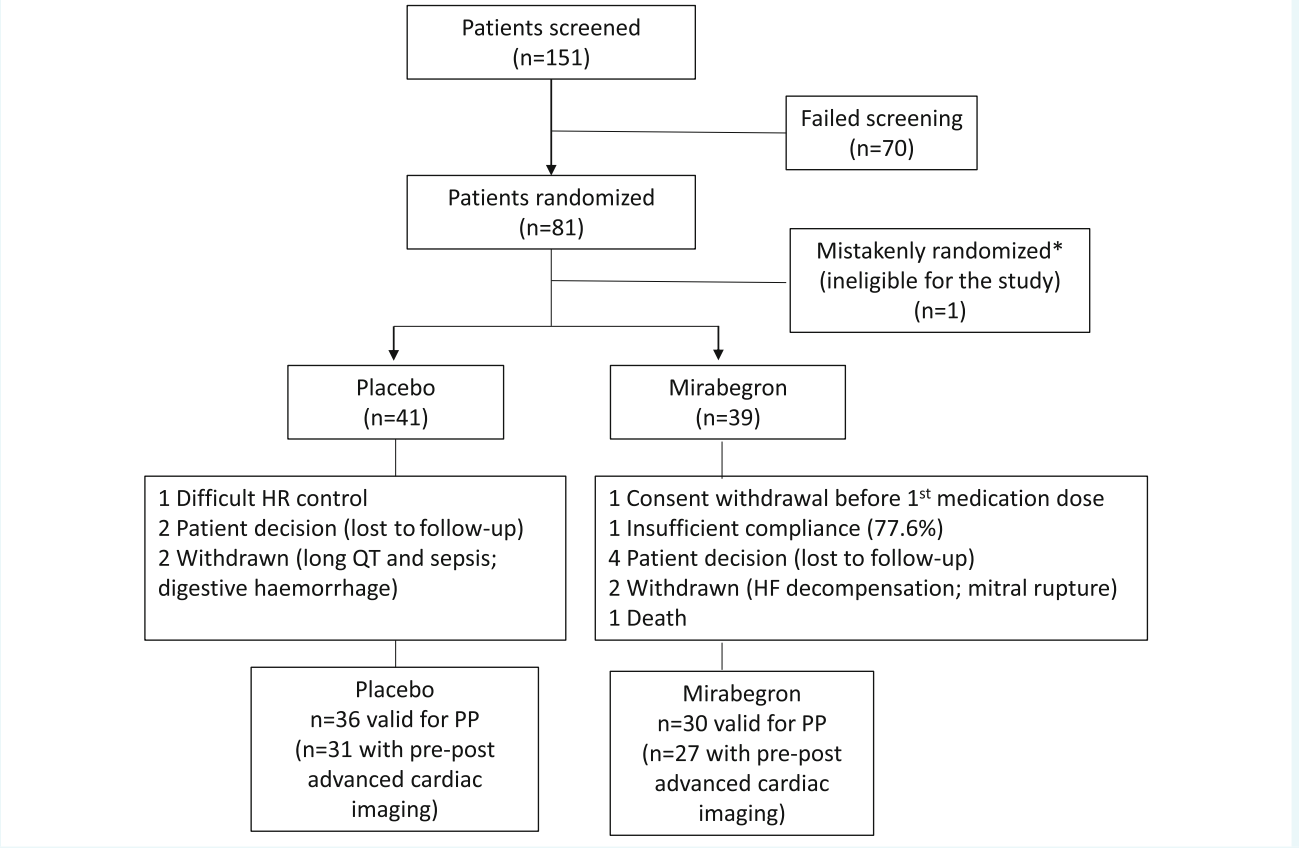


Figure 2 Patients flow chart. Screening and randomization. *One patient was mistakenly randomized because did not fulfil the inclusion criteria as she suffered from thromboembolic pulmonary hypertension. HF, heart failure; HR, heart rate; PP, per protocol population.

and no patient had severe degenerative valve disease at inclusion. In concordance with the study protocol, patients were receiving state-of-the-art medical and/or device therapy at baseline. In HFpEF, optimized evidence-based pharmacological treatment was focused to the optimal treatment of comorbidities (hypertension, diabetes, heart rate control in the case of atrial fibrillation) and hypervolaemia with the use of diuretics. Most patients were treated with beta-blockers, being selective beta-1 blockers the majority (bisoprolol [$n = 32$, 48%], carvedilol [$n = 10$, 15%], atenolol [$n = 5$; 7.5%], nebivolol [$n = 1$, 1.5%], and sotalol [$n = 1$, 1.5%]). Median therapeutic compliance was 100% of prescribed doses in both groups and median achieved dose was 150 mg in the mirabegron and 125 mg in the placebo group. Of the 66 patients who constituted the PP cohort (with pre-post evaluation of the primary outcome), 58 also had pre-post evaluable advanced cardiac imaging. At baseline, all except two patients underwent CMR ($n = 42$) or CCT ($n = 22$), whereas all but other two had CMR or CCT performed at 16-week follow-up ($n = 37$ and $n = 27$, respectively). Reasons for not performing advanced imaging in these four patients were technical problems ($n = 3$) and patient fragility due to recent hospital admission ($n = 1$). In another four cases, images were considered to be of insufficient quality to be evaluable by the Imaging Core Lab.

Primary and secondary outcomes

The primary outcome, placebo-corrected change from baseline to week 16 in PVR, was not met since there was a median change of +0.09 WU (Q1−Q3, −0.49, +1.05 WU) in the mirabegron arm and a median change of −0.71 WU (Q1−Q3, −1.46, +0.84 WU) in the placebo arm (placebo-corrected difference in PVR of 0.62 WU; 95% confidence interval [CI] −0.38, 1.61; $p = 0.218$). A similar result was obtained in the ITT population (placebo-corrected difference in PVR of 0.63 WU; 95% CI −0.22, 1.51; $p = 0.152$). *Table 3* shows the effect of mirabegron on the primary and pre-specified secondary outcomes. The study resulted positive for the pre-specified secondary outcome RV ejection fraction by advanced cardiac imaging (placebo-corrected change from baseline to week 16 of 3.0%; 95% CI 0.4, 5.7%; $p = 0.026$). No significant differences were observed between groups in the rest of pre-specified secondary outcomes.

Pre-specified subgroups analysis

Pre-specified analysis of subgroups according to baseline left ventricular ejection fraction (<40% vs. ≥40%) and maximal tolerated dose were performed for the primary outcome. The interaction terms between dichotomized left ventricular ejection fraction

Table 2 Baseline demographic and clinical characteristics according to treatment allocation

	Mirabegron ($n = 30$)	Placebo ($n = 36$)
Age (years)	69.7 ± 9.2	64.8 ± 10.6
Male sex	12 (40.0)	17 (47.2)
Diabetes mellitus	14 (46.7)	18 (50.0)
Atrial fibrillation	22 (73.3)	28 (77.8)
Heart disease		
Ischaemic	9 (30.0)	10 (27.8)
Non-ischaemic	21 (70)	26 (72.2)
HFpEF	21 (70)	26 (72.2)
Left ventricular ejection fraction (%)	50.7 ± 14.5	51.7 ± 15.6
NYHA functional class		
II	14 (46.7)	23 (63.9)
III	16 (53.3)	13 (36.1)
Previous non-coronary cardiac surgery	9 (30.0)	10 (27.8)
Pacemaker	2 (6.7)	4 (11.1)
Cardiac resynchronization therapy	2 (6.7)	0 (0.0)
Defibrillator	7 (23.3)	11 (30.6)
Beta-blockers	22 (73.3)	27 (75)
ACEI, ARB or ARNI	22 (73.3)	24 (66.6)
Mineralocorticoid receptor antagonists	14 (46.6)	22 (61.1)
Loop diuretics	28 (93.3)	32 (88.9)
Thiazide diuretics	3 (10.0)	2 (5.6)
Dihydropyridine calcium channel blockers	2 (6.7)	2 (5.6)
Ivabradine	5 (16.7)	1 (2.8)
Amiodarone	1 (3.3)	5 (13.9)
Oral anticoagulants	22 (73.3)	29 (80.6)

Data are presented as mean ± standard deviation, or *n* (%).
ACEI, angiotensin-converting enzyme inhibitor; ARB, angiotensin II receptor blocker; ARNI, angiotensin receptor–neprilysin inhibitor; HFpEF, heart failure with preserved ejection fraction; NYHA, New York Heart Association.

(*Table 4*) and treatment or maximal tolerated dose (as an ordinal variable) and treatment were not significant (*p*-values of 0.513 and 0.340, respectively). Similar results were obtained using the cut-off of 50% (data not shown). The effect of mirabegron after adjustment by left ventricular ejection fraction remained not statistically significant (0.52 WU; 95% CI −0.5, 1.54 WU) and neither after adjustment by maximal tolerated dose (0.62 WU, 95% CI −0.37, 1.6 WU). An exploratory (not pre-specified) analysis did not show significant differences in treatment effect in patients in sinus rhythm or atrial fibrillation (data not shown).

Other variables

Online supplementary *Table S1* displays symptoms, physical examination and variables monitored during the 6-min walking test, at baseline and 16-week follow-up. The percentage of patients presenting genitourinary symptoms or headache, typically associated with β3AR agonists, was very low. Mirabegron seemed not to affect heart rate, systolic blood pressure or oxygen saturation either at rest or after exercise. The effect of mirabegron on other (not included in the primary and secondary outcomes) systemic and pulmonary haemodynamics evaluated by right heart catheterization was neutral (online supplementary *Table S2*). Online supplementary *Table S3* shows cardiac advanced imaging parameters not included in the primary or secondary outcomes and measures obtained by echocardiography. Globally, there were no robust differences in echocardiographic parameters between groups, except for a significant reduction in left atrial diameter ($p = 0.016$) and a trend towards reduction of RV wall thickness ($p = 0.056$) in the group of mirabegron. Regarding the association between imaging techniques, at baseline and 16-week evaluations, RV ejection fraction by CMR/CT significantly correlated with tricuspid annular plane systolic excursion, tricuspid S wave velocity, RV fractional area change and RV global longitudinal strain ($p < 0.002$ for Pearson correlation in all cases). The highest correlation was found for RV fractional area change ($R = 0.75$ at baseline and $R = 0.66$ at follow-up), as previously observed.[22] Changes in RV ejection fraction by CMR/CT significantly correlated with changes in RV fractional area change ($R = 0.3$, $p = 0.023$).

Safety measures

A total of 238 AEs were reported, 119 in the mirabegron and 119 in the placebo group, regarding 74 patients (38 in the mirabegron and 36 in the placebo group) (*Table 5*). Severe AEs were reported in 11 patients in the mirabegron group and 9 in the placebo group. Of them, six events were considered to be related with the study medication by the independent data safety monitoring board, three in the mirabegron (chest pain in one patient, and incident atrial fibrillation and aggravated HF in another patient); and three in the placebo group (aggravated HF, incident atrial fibrillation and urinary tract infection). Study medication was discontinued because of serious AEs in two patients in the mirabegron group (HF decompensation and evidence of mitral rupture requiring surgery) and two patients in the placebo group (long QT in the context of sepsis, and a gastrointestinal bleeding). There were two deaths during the study, both in patients allocated to mirabegron. One patient presented a sudden cardiac death despite wearing an implantable automatic defibrillator. The second patient had been withdrawn from the study because of the presence of a mitral rupture requiring surgery and died in the postoperative period. None of them were considered to be related with the study drug. As shown in online supplementary *Table S4*, no clinically relevant changes were seen in vital signs, ECG or laboratory variables.

Discussion

SPHERE-HF is the first study to assess the effect of a β3AR agonist in patients with PH secondary to LHD. The primary endpoint, placebo-corrected change from baseline to week 16 in PVR, was not met. Consequently, interpretation of secondary endpoints

Table 3 Effect of mirabegron on primary and secondary outcomes

	Mirabegron ($n = 30$)	Placebo ($n = 36$)	Placebo-corrected mean difference (95% CI)	p-value
Primary outcome				
PVR (WU)				
Baseline	4.00 (3.42–4.62)	4.66 (3.75–6.23)	0.62 (−0.38, 1.61)	0.218
Week 16	4.09 (3.00–5.88)	4.54 (2.23–6.14)		
Secondary outcomes				
RV ejection fraction (%)				
Baseline	44.9 ± 10.1	45.5 ± 9.3	3.0 (0.4, 5.7)	0.026
Week 16	48.3 ± 8.8	45.0 ± 9.6		
Mean PAP (mmHg)				
Baseline	40.2 ± 9.6	44.0 ± 10.1	−0.5 (−4.2, 3.3)	0.812
Week 16	38.3 ± 10.6	40.9 ± 8.2		
TPG (mmHg)				
Baseline	16.0 (14.0–21.5)	19.0 (15.8, 28.0)	2.1 (−1.0, 5.2)	0.206
Week 16	15.6 (12.0–19.8)	18.0 (13.8–23.2)		
CO (L/min)				
Baseline	4.2 ± 1.2	4.4 ± 1.3	−0.2 (−0.6, 0.2)	0.391
Week 16	4.0 ± 1.1	4.3 ± 1.3		
Diastolic gradient (mmHg)				
Baseline	2.0 (0.0–5.8)	5.5 (1.8–8.2)	1.1 (−1.3, 3.3)	0.385
Week 16	3.0 (0.0–5.0)	3.0 (1.0–8.2)		
NYHA functional class				
Baseline			OR 0.34 (0.09–1.08)	0.077
II	14 (46.7)	21 (58.3)		
III	16 (53.3)	15 (41.7)		
Week 16				
I	3 (10.0)	4 (11.1)		
II	15 (50.0)	24 (66.7)		
III	12 (40.0)	8 (22.2)		
Quality of life (KCCQ overall summary score) (points)				
Baseline	58.8 ± 21.3	64.9 ± 18.9	−7.9 (−16.4, 0.6)	0.067
Week 16	60.3 ± 20.2	70.6 ± 20.7		
6MWT distance (m)				
Baseline	357 (270–399)	368 (322–444)	5.3 (−25.1, 35.8)	0.720
Week 16	340 (281–414)	370 (314–434)		
Borg scale (points)				
Baseline	3.0 (1.0–5.0)	3.0 (1.2–4.0)	−0.4 (−1.7, 0.9)	0.581
Week 16	2.0 (1.0–4.0)	3.0 (1.2–4.8)		
NT-proBNP (pmol/L)				
Baseline	1391 (802–3106)	1750 (1140–2485)	−92 (−562, 364)	0.703
Week 16	1151 (741–2540)	1743 (1057–2645)		

Data are presented as mean ± standard deviation, *n* (%), or median (first–third quartile).
6MWT, 6-min walking test; CI, confidence interval; CO, cardiac output; KCCQ, Kansas City Cardiomyopathy Questionnaire; NT-proBNP, N-terminal pro-B-type natriuretic peptide; OR, odds ratio; PAP, pulmonary arterial pressure; PVR, pulmonary vascular resistance; RV, right ventricular; TPG, transpulmonary gradient; WU, Wood units.

requires caution. In relation to these, patients receiving mirabegron presented a significantly better evolution of RV ejection fraction during the course of the trial as compared to placebo without other significant differences in the rest of pre-specified secondary outcomes. Importantly, mirabegron was well tolerated and had a favourable safety profile (*Graphical Abstract*).

Treatment of PH in patients with HF remains an unmet clinical need due to its high prevalence and negative prognostic impact. Although different pharmacological treatment options are available in patients with pulmonary arterial hypertension (PAH), none of them has shown a beneficial effect in PH-LHD.[23] Randomized clinical trials evaluating the effect of prostanoids and bosentan in patients with advanced HF and left ventricular systolic dysfunction were stopped due to an increase in AEs.[24–26] While these initial clinical trials did not select the population based on the presence or absence of PH, the subsequent results of the MELODY

Table 4 Pre-specified subgroup analysis

	Mirabegron (n = 30)		Placebo (n = 36)		p-value interaction
	LVEF ≥40% (n = 21)	LVEF <40% (n = 9)	LVEF ≥40% (n = 26)	LVEF <40% (n = 10)	
PVR					0.513
Baseline	5.05 (2.46)	3.77 (0.90)	5.37 (2.36)	4.66 (1.83)	
Week 16	4.91 (3.09)	4.98 (3.14)	4.87 (1.99)	4.68 (3.37)	

LVEF, left ventricular ejection fraction; PVR, pulmonary vascular resistance.

Table 5 Adverse events

	Mirabegron (n = 39)		Placebo (n = 41)	
	No. of SAEs	No. of patients with AEs (%)	No of SAEs	No of patients with AEs (%)
Safety endpoint				
Death	2		0	
Hospitalization or need for i.v. furosemide due to heart failure	6	5 (13)	3	3 (7)
Investigator-reported adverse event				
SAEs	17	11 (28)	14	9 (22)
SAE possibly related to the study drug	3	2 (5)	3	3 (7)
Any AE	119	38 (97)	119	36 (88)
Cardiac				
Heart failure, dyspnoea or oedema	23	16 (41)	17	15 (37)
Incident or decompensated atrial flutter or fibrillation	3	3 (8)	3	3 (7)
Ventricular arrhythmias	2	2 (5)	1	1 (2)
QT interval prolongation	2	2 (5)	4	4 (10)
Respiratory				
Respiratory tract infections	24	15 (38)	18	13 (32)
Urinary				
Urinary symptoms or infection	3	3 (8)	8	7 (17)
Nervous system				
Headache	4	3 (8)	2	2 (5)

AE, adverse event; i.v., intravenous; SAE, serious adverse event.

clinical trial with macitentan[27] focused on patients with CpcPH, reinforced the message of lack of benefit compared to placebo. Phosphodiesterase 5 inhibitors have also failed to demonstrate a beneficial effect in PH-LHD patients. Within this group, in PH-LHD secondary to HFrEF, there is only one small single-centre study published by Lewis *et al.*[28] in which sildenafil improved exercise capacity and quality of life associated with a significant reduction in PVR, and some observational studies that have reported haemodynamic improvement prior to heart transplantation.[29] In HFpEF, the randomized study by Guazzi *et al.*[30] using sildenafil showed encouraging results; however, these were not replicated in the RELAX trial[31] or in the randomized trial by Hoendermis *et al.*,[32] that failed to show beneficial effects, and neither in the multicentre clinical trial SIOVAC.[33] Indeed, in this latest trial, which included 200 patients with residual PH after valve surgery, the sildenafil-treated group demonstrated a higher rate of AEs. Finally, riociguat, a soluble guanylate cyclase stimulator, did not improve the primary outcome (mean PAP) both in PH secondary to HFrEF[34] and HFpEF.[35] In this regard, the consensus of the 6th World Symposium on Pulmonary Hypertension[36] and recently published 2022 ESC/ERS guidelines[6] included a strong recommendation against the use of pulmonary vasodilators in PH-LHD, particularly in isolated post-capillary PH-LHD. Regarding riociguat, a very recent trial has reported a significant increase in the cardiac output (in this case defined as the main outcome) in patients with PH associated with HFpEF.[37] Of note, most of the aforementioned trials lacked a comprehensive evaluation of RV function by cardiac imaging.

Mirabegron is a novel class of drugs that act stimulating β3AR. Like other adrenoceptors, β3ARs are coupled to G proteins and the downstream pathway after activation includes nitric oxide (NO) synthase, NO-activated guanylyl cyclase and cyclic guanosine and adenosine monophosphate synthesis.[37] Loss of cyclic guanosine and adenosine monophosphates is a hallmark of PH.

Within pulmonary circulation, cyclic nucleotides are responsible for mediating endothelin-dependent dilatation, thereby maintaining pulmonary vascular homeostasis, but they also have valuable actions on pulmonary vascular remodelling, fibrosis and RV function.[38] Because of its promising pre-clinical results in a swine model of post-capillary PH[14] and the safe profile demonstrated in patients with overactive bladder syndrome and HF,[15–18] we considered that mirabegron deserved to be evaluated in patients with PH secondary to HF.

SPHERE-HF is the first phase II trial exploring the effect of a β3AR in patients with CpcPH secondary to HFrEF or HFpEF. We chose this population because we share the opinion[20] that it is the added pre-capillary component which has the greatest impact on prognosis in group 2 PH. Although we recognize that pathophysiology of HFrEF and HFpEF differs, we considered that CpcPH selects a cohort of patients who share a common phenotype with severe pulmonary remodelling and RV dysfunction,[20] and therefore most likely to benefit from a targeted therapy. The design of the trial coincided with the publication of the 2015 ESC/ERS guidelines.[23] The reason for using PVR, diastolic pressure and transpulmonary gradient in the discrimination of CpcPH was that transpulmonary gradient was widely used in clinical practice at that moment and diastolic pressure had been recently incorporated in ESC/ERC guidelines. However, all patients fulfill the current definition of CpcPH based on the recently published 2022 ESC/ERS guidelines[6] and only six patients do not fulfill definition of CpcPH according to the 2018 World Symposium[27] (three randomized to mirabegron and three to placebo). At inclusion, PVR severity was lower than in the trial by Guazzi *et al.*[30] in HFpEF (significant reduction of mean PAP using sildenafil), and slightly higher compared to the SIOVAC trial[33] in PH associated with corrected valve disease (negative results using sildenafil) and the recently published DYNAMIC trial in HFpEF[36] (significant improvement of cardiac output with vericiguat). We decided to use the change in PVR as the primary outcome as the most robust haemodynamic parameter in this population.[39,40] Selecting endpoints for phase II clinical trials in PH is not straightforward and have relevant implications, for example, as discussed in the editorial comment by Bauersachs and Olsson,[41] the clinical relevance of the positive result of the DYNAMIC trial using cardiac output as the endpoint is unclear. However, we acknowledged that RV systolic function has even a higher prognostic impact than haemodynamic severity in group 2 PH[1]. Moreover, it is not uncommon that both variables vary in a non-coordinated way. Thereby, PAH patients may show a progressive decline in RV function despite a therapeutic response to vasodilators.[42]

In our study, compared to patients assigned to placebo, patients receiving mirabegron failed to achieve a significant reduction in PVR after 16 weeks of treatment. In addition, in the pre-specified subgroup analysis we found no evidence of a differential effect of mirabegron on PVR in the HFrEF or HFpEF populations. In fact, pulmonary haemodynamics, including systolic and mean PAP, PAWP and cardiac output, as assessed by RHC, remained virtually steady between the baseline and 16-week evaluations in both groups. These results differ from those recently reported by Bundgaard *et al.*[43] in which patients with NYHA functional class III–IV HFrEF receiving mirabegron (300 mg) for 7 days presented a significant increase in cardiac output and reduction of PVR, compared to placebo. This discrepancy may relay in differences between trials in patients' characteristics (left ventricular ejection fraction of 26 ± 7% vs. 48 ± 17%, NYHA functional class ≥III in all patients vs. 44% in ours), duration (7 days vs. 16 weeks) and dose (300 mg vs. median of 150 mg). In contrast, mirabegron was associated with a significantly better evolution of RV ejection fraction assessed by cardiac advanced imaging, in the absence of significant differences in systemic blood pressure or heart rate. Clinical experience and previous studies have shown that PVR and RV ejection fraction may respond to PH therapies in a different manner. Thus, in the study by van der Veerdonk *et al.*,[42] PAH patients receiving PAH-targeted therapies presented a significantly reduction in PVR and a significantly increase in cardiac output but not differences in RV ejection fraction. Several mechanisms have been proposed to play a role in RV ejection fraction besides afterload.[42,44] Regarding the clinical relevance that a 3% increase of RV ejection fraction could have, a recent meta-analysis published by Alabed *et al.*[45] demonstrated that a 1% reduction in RV ejection fraction in patients with PAH was associated with a 4.9% increase in the risk of clinical worsening and a 2.1% increase in the risk of death. To our knowledge there are no similar quantitative data for CpcPH, other than the demonstrated independent and additive prognostic value of RV ejection fraction.[1] The indistinct use of CMR or CCT for the measurement of RV ejection fraction is justified by previous literature that demonstrates an extraordinarily high concordance between both techniques, with a correlation coefficient of 0.98 and a pooled standard mean difference of 0.65 (95% CI −2.60, 1.29) in a recent meta-analysis.[46] However, beyond a reduction in left atrial diameter and a trend towards a reduction in RV wall thickness, no robust changes were observed in the echocardiographic parameters. In this regard, it is known that the statistical power to assess changes in cardiac remodelling and function using CMR is significantly higher than using echocardiography[47] and therefore it could be plausible that an eventual beneficial effect of mirabegron was noted by CMR or CCT and not by echocardiography. In fact, changes in RV ejection fraction by CMR significantly correlated with changes in RV fractional area change. If real, this beneficial effect would be independent of pressure afterload relief and could be associated with the cardioprotective effect observed in previous studies of ischemia–reperfusion[12,48,49] and HF[11,50,51] that involved an improvement in myocardial perfusion, antioxidant and antifibrotic effects. In this sense, Bundgaard *et al.*[50] demonstrated that the β3AR stimulation produced an increased cardiac contractility in sheep after induction of HF that did not occur before induction of the model (healthy myocardium). Protection of the alpha1 subunit of the Na^+/K^+ ATPase from inactivation by oxidative S-glutathionylation and subsequent reduction of intracellular Na^+ content has been considered critical for the improvement of inotropism in the context of HF.[50] In addition, β3AR may regulate cardiac function through paracrine effects from NO- and endothelium-derived hyperpolarizing factor-dependent vasorelaxation of coronary arteries.[8] Finally, demonstrated coupling of β3AR to cGMP and downstream activation of PKG-dependent signalling may result in inhibition of hypertrophy.[52] A direct inotropic

effect attributed to off-target stimulation is unlikely due to the mirabegron's high selectivity towards β3AR, the high percentage of patients under beta-blockers and the absence of significant changes in heart rate or systemic blood pressure. Nevertheless, this eventual beneficial effect observed in RV ejection fraction should be evaluated with caution as it was a secondary outcome, available for the 88% of the cohort, and was not associated with a significant improvement in functional class or quality of life. In addition, although there were no differences in the number and severity of AEs, a higher number of patients in the mirabegron group withdrew study medication and there were two patients who died (though considered not attributable to study medication by the safety external committee).

The present study was designed and developed with a robust methodology (multicentre, randomized, double-blind clinical trial), however we can point out some limitations. It included a heterogeneous cohort of patients with both HFrEF and HFpEF. Despite being aware of the differences in the pathophysiology of both entities, we prioritized the fact that all patients had CpcPH, as we hypothesized that the main effect exerted by mirabegron would be the reduction of PVR. Considering that the beneficial effect might be greater at the myocardial level, it would possibly have been more appropriate to focus on a single HF phenotype. Measurement of cardiac output by RHC, needed for PVR calculation, was done by thermodilution, which may be inaccurate in patients with severe tricuspid regurgitation (present in 10 patients at baseline and 11 at 16-week follow-up). This fact may explain, at least partially, the discordance observed between the increase in RV ejection fraction and the absence of a significant increase in cardiac output. Indeed, in the subpopulation with available advanced imaging, cardiac output by CMR did increase in patients receiving mirabegron (though differences did not reach significance as compared to placebo). Two thirds of patients had atrial fibrillation at baseline, and we cannot exclude a lower benefit in these patients compared to those in sinus rhythm, as has been observed with the use of beta-blockers in HFrEF,[53] although the results of the exploratory analysis do not point in this direction. Also, approximately one third had PH associated with corrected valve disease, subgroup probably representing long evolution chronic PH, especially refractory to treatment. Another limitation is associated with the baseline observation carried forward approach used to deal with missing data in the ITT population, which assumes no changes in patients receiving placebo or mirabegron. Nevertheless, no differences were observed in the results obtained in the PP or the ITT population.

Conclusions

The SPHERE-HF trial, the first to evaluate the safety and efficacy of mirabegron in CpcPH, did not achieve the primary outcome, since the placebo-corrected change from baseline to week 16 in PVR was not met. On pre-specified secondary outcomes, a significant improvement in RV ejection fraction assessed by advanced cardiac imaging was found without differences in functional class or quality of life.

Supplementary Information

Additional supporting information may be found online in the Supporting Information section at the end of the article.

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Conflict of interest: CNIC and Fundació Clínic per a la recerca biomèdica hold a patent for the use of beta-3 agonists for the treatment of pulmonary hypertension (B.I., A.G.A., and V.F. are co-inventors). All other authors have nothing to disclose.

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