Supplementary appendix

TABLE S1 Inclusion and exclusion criteria

Inclusion criteria
1. Written informed consent prior to initiation of any study-mandated procedure
2. Males or females ≥18 years of age
3. Chronic heart failure fulfilling all of the following criteria: - Relevant structural heart disease and/or diastolic dysfunction[1] - Ejection fraction ≥30% measured by echocardiography - NYHA FC II or III
4. PH in WHO group 2 (NICE classification[2]) with a PAWP >15 mmHg, confirmed by any previous right heart catheterisation
5. Right heart catheterisation within 12 weeks prior to the Screening visit or during the Screening period with the following data: - mPAP ≥25 mmHg at rest - PAWP or LVEDP <25 mmHg - PVR at rest ≥240 dyn·sec/cm^5 - DPG ≥7 mmHg
6. Optimal treatment of left heart failure with stable dose of oral diuretic(s) (and other standard heart failure treatment, if applicable) for at least 1 week prior to baseline right heart catheterisation and up to Randomisation
7. 6-minute walk distance ≥150 m at Screening or Randomisation
8. For women of child-bearing potential, it was necessary to have a negative pre-treatment serum pregnancy test, agreement to undertake monthly serum pregnancy tests, and agreement to use accepted methods of birth control from Screening to last 30 days after study treatment discontinuation
Exclusion criteria
1. PH associated with significant unrepaired structural valvular heart disease (*i.e.* moderate or severe aortic stenosis, aortic insufficiency, mitral stenosis, mitral regurgitation still persisting after fluid control). Repaired valvular heart disease and valvular replacement allowed.

2. Evidence of pulmonary oedema
3. Presence of orthopnoea
4. Presence of moderate-to-severe peripheral oedema despite optimal diuretic therapy
5. History of pulmonary thromboembolism with segmental defect on ventilation/perfusion scan
6. Systemic hypertension, *i.e.* SBP >180 mmHg or DBP >100 mmHg
7. SBP <90 mmHg
8. Uncontrolled heart rate from atrial fibrillation (>100 beats per minute)
9. Clinically known pulmonary veno-occlusive disease
10. Untreated obstructive sleep apnoea
11. Unstable coronary artery disease
12. Severe hepatic impairment
13. Myocardial infarction within 6 months prior to Randomisation
14. Obstructive, restrictive, and infiltrative cardiomyopathies, *e.g.* amyloidosis
15. Severe obstructive lung disease defined as FEV_1/FVC <0.7 associated with FEV_1 <50% of predicted value after bronchodilator administration
16. Known severe emphysema, *e.g.* total lung capacity >120% of predicted value
17. Known moderate-to-severe restrictive lung disease
18. PaO_2 <60 mmHg or SpO_2 <90% at room air
19. Estimated creatinine clearance <30 mL/min
20. AST and/or ALT >3×ULN at Screening
21. Haemoglobin <100 g/L at Screening
22. Pregnancy, or planned to become pregnant or lactating
23. Administration of PAH-specific therapy (*i.e.* ERAs, prostanoids, PDE-5 inhibitors, guanylate cyclase stimulators) or another investigational drug in the 1-month period prior to baseline right heart catheterisation
24. Treatment with strong cytochrome P-450 3A4 (CYP3A4) inducers, such as rifabutin, rifampin, rifampicin, rifapentin, carbamazepine, phenobarbital, phenytoin, or St John's wort, in the 1-month period prior to Randomisation
25. Known hypersensitivity to drugs of the same class as the study treatment (*i.e.* ERAs), or to any of

the study treatment excipients
26. Any condition that prevented compliance with the protocol or adherence to therapy
27. Previous right heart catheterisation with severe complications, such as (but not limited to) cardiac arrest, arrhythmia requiring intervention, pulmonary haemorrhage, stroke, thromboembolic event, and pulmonary hypertensive crisis

ALT, alanine aminotransferase; AST, aspartate aminotransferase; DBP, diastolic blood pressure; DPG, diastolic pressure gradient; ERA, endothelin receptor antagonist; FEV_1, forced expiratory volume in 1 second; FVC, forced vital capacity; LVEDP, left ventricular end diastolic pressure; mPAP, mean pulmonary arterial pressure; NICE, National Institute for Health and Care Excellence; NYHA FC, New York Heart Association Functional Class; PAH, pulmonary arterial hypertension; PaO_2, partial pressure of oxygen in atrial blood; PAWP, pulmonary arterial wedge pressure; PDE-5, phosphodiesterase type-5; PH, pulmonary hypertension; PVR, pulmonary vascular resistance; SBP, systolic blood pressure; SpO_2, saturation of peripheral oxygen; ULN, upper limit of normal; WHO, World Health Organization.

TABLE S2 Treatment-emergent serious adverse events

n (%)	Macitentan n=31	Placebo n=32
Patients with ≥1 serious adverse event	11 (35.5)	6 (18.8)
Pneumonia	2 (6.5)	0
Cardiac failure acute	1 (3.2)	0
Cardiac failure congestive	1 (3.2)	1 (3.1)
Cardiorenal syndrome	1 (3.2)	0
Chronic respiratory failure	1 (3.2)	0
Hypoglycaemia	1 (3.2)	0
Jugular vein thrombosis	1 (3.2)	0
Left ventricular failure	1 (3.2)	0
Mouth haemorrhage	1 (3.2)	0
Oedema peripheral	1 (3.2)	1 (3.1)
Respiratory failure	1 (3.2)	0
Pulmonary mass	1 (3.2)	0
Pulmonary oedema	1 (3.2)	0
Pulmonary congestion	1 (3.2)	0
Respiratory tract infection bacterial	1 (3.2)	0
Right ventricular failure	1 (3.2)	1 (3.1)
Sudden death	1 (3.2)	0
Ventricular tachycardia	1 (3.2)	0
Acute kidney injury	0	1 (3.1)
Atrial fibrillation	0	1 (3.1)
Fall	0	1 (3.1)

TABLE S3 Adverse events leading to study treatment discontinuation

n (%)	Macitentan n=31	Placebo n=32
Patients with ≥1 adverse event leading to discontinuation	5 (16.1)	0
Dyspnoea	1 (3.2)	0
Respiratory failure	1 (3.2)	0
Right ventricular failure	1 (3.2)	0
Mouth haemorrhage	1 (3.2)	0
Jugular vein thrombosis	1 (3.2)	0

TABLE S4 Adverse events of special interest

	Macitentan n=31	Placebo n=32
Related to oedema and fluid overload		
Patients with ≥1 adverse event	8 (25.8)	6 (18.8)
Oedema/peripheral oedema	4 (12.9)	4 (12.5)
Fluid retention/overload	3 (9.7)	0
Pulmonary congestion/oedema	2 (6.4)	0
Pleural effusion	0	2 (6.3)
Related to anaemia		
Patients with ≥1 adverse event	4 (12.9)	1 (3.1)
Anaemia	2 (6.5)	0
Iron-deficiency anaemia	0	1 (3.1)
Haemoglobin decreased	2 (6.5)	0

TABLE S5 Marked abnormalities in laboratory values*

Laboratory abnormality, n/N (%)	Macitentan n=31	Placebo n=32
Haemoglobin		
LL† (<100 g/L)	5/28 (17.9)	2/32 (6.3)
Haematocrit		
HH (>55% for females, >60% for males)	1/28 (3.6)	0/32
Lymphocytes		
LL (<0.8×10^9/L)	4/28 (14.3)	1/32 (3.1)
LLL (<0.5×10^9/L)	2/28 (7.1)	0/32
HH (<4.0×10^9/L)	0/28	1/32 (3.1)
Platelets		
LL (<75×10^9/L)	1/28 (3.6)	0/32
Alkaline phosphatase		
HH (2.5×ULN)	0/28	1/32 (3.1)
Bilirubin		
HH (>2×ULN)	2/28 (7.1)	0/32
Creatinine‡		
HH (>1.5×baseline or 1.5×ULN)	8/29 (27.6)	5/32 (15.6)
Blood urea nitrogen		
HH (>2.5×ULN)	4/29 (13.8)	3/32 (9.4)
Urate		
HH (>0.59 mmol/L)	1/28 (3.6)	2/32 (6.3)
HHH (>0.72 mmol/L)	1/28 (3.6)	0/32
Glucose		
LL (<3.0 mmol/L)	0/29	1/32 (3.1)
HH (>8.9 mmol/L)	4/29 (13.8)	3/32 (9.4)
HHH (>13.9 mmol/L)	1/29 (3.4)	1/32 (3.1)
Sodium		

LLL (<130 mmol/L)	1/29 (3.4)	1/32 (3.1)
Potassium		
LL (<3.2 mmol/L)	2/29 (6.9)	1/32 (3.1)
LLL (<3.0 mmol/L)	1/29 (3.4)	0/32
HH (>5.5 mmol/L)	5/29 (17.2)	1/32 (3.1)
HHH (>6.0 mmol/L)	2/29 (6.9)	0/32
Magnesium		
HHH (>1.23 mmol/L)	1/28 (3.6)	1/32 (3.1)

HH/HHH, marked high abnormality; LL/LLL, marked low abnormality; n, number of patients; N, number of patients with available data; ULN, upper limit of normal. *Frequencies represent the number of patients with the defined abnormality reported at least once; †No patients reported haemoglobin values <80 g/L. In four of the five macitentan-treated patients, the magnitude of decrease from baseline was ≥20 g/L. In all cases, haemoglobin levels returned to normal or to near baseline levels by end of study. In two of the macitentan-treated patients, the decrease from baseline was ≥50 g/L. In both cases, haemoglobin returned to a value close to normal by end of study; ‡Most patients with marked abnormalities in creatinine had elevated creatinine levels at baseline.

TABLE S6 Change in haemodynamic parameters from baseline to Week 12

	Macitentan				Placebo				
	n	**Baseline**	**Week 12**	**Change**	**n**	**Baseline**	**Week 12**	**Change**	**Treatment effect at Week 12**
Parameter, unit		**Mean (SD)**		**Geometric mean (95% CL)***		**Mean (SD)**		**Geometric mean (95% CL)***	**Ratio of geometric means (95% CL)†**
PVR, dyn·sec/cm^5	20	525.5 (270.9)	357.2 (195.0)	66.3 (56.2–78.3)	24	521.4 (233.9)	438.7 (255.4)	71.2 (51.4–98.8)	0.93 (0.64–1.36)
	Macitentan				**Placebo**				
	n	**Baseline**	**Week 12**	**Change**	**n**	**Baseline**	**Week 12**	**Change**	**Treatment effect at Week 12**
			Mean (SD)				**Mean (SD)**		**Mean absolute change from baseline (95% CL)**

mPAP, mmHg	21	44.6 (8.7)	41.1 (10.7)	-3.5 (5.7)	25	45.9 (10.2)	42.1 (11.2)	-3.8 (9.1)	0.3 (-4.3–4.9)
mRAP, mmHg	21	12.1 (4.6)	11.2 (5.7)	-0.9 (5.7)	25	13.0 (4.7)	11.3 (5.2)	-1.6 (4.0)	0.7 (-2.2–3.6)
PAWP, mmHg	20	19.1 (3.6)	19.9 (7.0)	0.8 (6.5)	24	19.7 (3.6)	20.8 (7.0)	1.1 (6.5)	-0.3 (-4.2–3.7)
TPR, dyn·sec/cm^5	21	889.7 (409.5)	671.7 (285.3)	-218.0 (264.7)	25	882.9 (368.6)	827.1 (361.0)	-55.8 (258.0)	-162.2 (-318.0–6.5)
Cardiac index, L/min/m^2	20	2.3 (0.6)	2.7 (0.7)	0.4 (0.5)	24	2.3 (0.6)	2.3 (0.6)	-0.0 (0.5)	0.4 (0.1– 0.7)
Cardiac output (L/min)	21	4.5 (1.3)	5.3 (1.4)	0.8 (0.9)	25	4.5 (1.2)	4.5 (1.3)	-0.0 (0.8)	0.8 (0.3–1.4)
TPG, mmHg	20	26.0 (7.7)	21.6 (8.5)	–4.4 (5.3)	24	26.9 (7.5)	21.8 (10.0)	-5.0 (8.6)	0.7 (-3.7–5.1)
DPG, mmHg	20	11.8 (5.9)	7.0 (6.1)	-4.8 (5.3)	24	11.4 (4.4)	7.0 (7.9)	-4.3 (7.6)	-0.4 (-4.5–.6)
Mixed venous oxygen saturation, %	17	65.8 (8.4)	67.4 (7.4)	1.6 (5.2)	16	61.3 (8.2)	63.3 (8.2)	2.0 (6.5)	-0.4 (-4.6–3.8)

CL: confidence limit; DPG: diastolic pulmonary gradient; mPAP: mean pulmonary arterial pressure; mRAP: mean right atrial pressure; PAWP: pulmonary arterial wedge pressure; PVR: pulmonary vascular resistance; SD: standard deviation; TPG: transpulmonary pressure gradient; TPR: total peripheral resistance. *Geometric mean (95% CL) of percent ratio Week 12/baseline; †Ratio of geometric means (macitentan/placebo).

TABLE S7 Change in echocardiographic parameters from baseline to Week 12

	Macitentan				Placebo				
	n	**Baseline**	**Week 12**	**Change**	**n**	**Baseline**	**Week 12**	**Change**	**Treatment effect at Week 12**
Parameter, unit		**Mean (SD)**				**Mean (SD)**			**Mean absolute change from baseline (95% CLs)**
E/e' (lateral), ratio	15	10.7 (4.1)	11.6 (3.8)	0.91 (2.9)	19	13.2 (7.9)	11.4 (5.1)	-1.78 (5.1)	2.69 (-0.32–5.70)
TAPSE, cm	21	1.6 (0.5)	1.8 (0.6)	0.16 (0.6)	21	1.6 (0.6)	1.6 (0.5)	-0.02 (0.6)	0.18 (-0.17–0.54)
Right ventricle fractional area change, %	23	40.0 (12.1)	37.4 (10.0)	-2.57 (14.7)	21	37.5 (10.9)	38.0 (9.7)	0.41 (7.3)	-2.98 (-10.16–4.19)
Right ventricular outflow tract acceleration time, msec	23	83.7 (17.2)	80.3 (23.8)	-3.48 (30.0)	25	91.0 (18.4)	88.5 (28.9)	-2.52 (26.0)	-0.96 (-17.23–15.32)
Right ventricular: left ventricular diastolic dimension ratio	23	0.9 (0.3)	0.9 (0.2)	-0.08 (0.2)	21	1.1 (0.2)	1.1 (0.2)	-0.01 (0.2)	-0.08 (-0.20–0.05)

Right ventricular: left ventricular diastolic area ratio	19	0.8 (0.2)	0.8 (0.3)	0.03 (0.2)	20	0.8 (0.3)	0.8 (0.4)	0.02 (0.3)	0.00 (-0.15–0.16)

CL: confidence limits; SD: standard deviation; TAPSE: tricuspid annular plane systolic excursion.

TABLE S8 Sensitivity analyses: Change in haemodynamic parameters from baseline to Week 12 after applying imputation rules for missing data

	Macitentan n=31			**Placebo n=32**			
Parameter, unit	**Baseline**	**Week 12**	**Change**	**Baseline**	**Week 12**	**Change**	**Treatment effect at Week 12**
	Mean (SD)		**Geometric mean (95% CL)***	**Mean (SD)**		**Geometric mean (95% CL)***	**Ratio of geometric means (95% CL)†**
PVR, dyn·sec/cm^5	491.2 (235.5)	388.7 (183.3)	77.7 (68.2–88.6)	549.3 (239.6)	487.3 (264.8)	77.5 (60.6–99.2)	1.00 (0.76–1.32)
	Baseline	**Week 12**	**Change**	**Baseline**	**Week 12**	**Change**	**Treatment effect at Week 12**
	Mean (SD)		**Absolute mean (SD)**	**Mean (SD)**		**Absolute mean (SD)**	**Mean absolute change from baseline (95% CL)**

mPAP, mmHg	46.4 (8.5)	45.3 (12.2)	-1.2 (6.4)	47.4 (9.9)	44.4 (11.2)	-3.0 (8.2)	1.8 (-1.9–5.5)
mRAP, mmHg	13.2 (4.6)	14.2 (8.0)	1.1 (6.7)	13.3 (4.8)	12.1 (5.3)	-1.3 (3.6)	2.3 (-0.4–5.0)
PAWP, mmHg	19.5 (3.3)	21.9 (7.9)	2.5 (7.2)	20.1 (3.9)	20.9 (6.5)	0.8 (5.6)	1.6 (-1.6–4.9)
TPR, dyn·sec/cm^5	844.5 (362.2)	695.8 (265.6)	-148.7 (239.1)	948.9 (365.0)	905.3 (367.9)	-43.6 (228.2)	-105.2 (-222.9–12.6)
Cardiac index, L/min/m^2	2.5 (0.7)	2.7 (0.7)	0.2 (0.5)	2.2 (0.6)	2.2 (0.6)	-0.03 (0.4)	0.21 (-0.01–0.44)
Cardiac output (L/min)	4.9 (1.5)	5.4 (1.4)	0.5 (0.9)	4.3 (1.1)	4.3 (1.2)	-0.02 (0.7)	0.52 (0.11–0.93)
TPG, mmHg	26.9 (7.5)	26.0 (11.9)	-1.0 (7.6)	27.3 (7.7)	23.6 (10.0)	-3.8 (7.7)	2.8 (-1.0–6.7)
DPG, mmHg	11.3 (6.1)	10.0 (10.3)	-1.3 (8.0)	11.1 (5.6)	7.8 (8.1)	-3.3 (6.8)	2.0 (-1.8–5.7)
Mixed venous oxygen saturation, %	67.7 (8.0)	67.5 (6.7)	-0.2 (5.7)	58.5(9.0)	59.9 (9.5)	1.4 (5.5)	-1.6 (-4.8–1.6)

CL: confidence limits; DPG: diastolic pulmonary gradient; mPAP: mean pulmonary arterial pressure; mRAP: mean right atrial pressure; PAWP: pulmonary arterial wedge pressure; PVR: pulmonary vascular resistance; SD: standard deviation; TPG: transpulmonary pressure gradient; TPR: total peripheral resistance. *Geometric mean (95% CL) of percent ratio Week 12/baseline; †Ratio of geometric means (macitentan/placebo). Imputation rules for missing data at Week 12: last observation carried forward for patients still alive and not hospitalised; for patients who died or were hospitalised for heart failure, the worst value was imputed.

MELODY-1 study investigators

The Macitentan in subjects with combined prE- and post-capiLlary pulmOnary hypertension due to left ventricular DYsfunction (MELODY) Investigators are as follows: Austria – D. Bonderman, R. Steringer-Mascherbauer; Belgium – M. Delcroix, J.L. Vachiéry; Canada – J. Swiston; Germany – S. Rosenkranz, J. Behr, M. Rosenberg; Czech Republic – P. Jansa, J. Spinar, M. Hutyra, H. Al-Hiti; France – F. Bauer; Israel – A. Yochai, N. Berkman, M. Segel, G. Fink; Italy – G. Sinagra; Spain – F. Pérez Villa, M.A. Gomez Sanchez, E. Domingo Ribas, J.M. Arizon del Prado; Switzerland – M. Schwerzmann, P. Yerly; USA – J.W. McConnell, S. Joseph, H.W. Farber, G. Ashrith, P.J. Engel.