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Effect of Phosphodiesterase-5 Inhibition on Exercise Capacity and Clinical Status in Heart Failure With Preserved Ejection Fraction

A Randomized Clinical Trial

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Importance Studies in experimental and human heart failure suggest that phosphodiesterase-5 inhibitors may enhance cardiovascular function and thus exercise capacity in heart failure with preserved ejection fraction (HFPEF).

Objective To determine the effect of the phosphodiesterase-5 inhibitor sildenafil compared with placebo on exercise capacity and clinical status in HFPEF.

Design Multicenter, double-blind, placebo-controlled, parallel-group, randomized clinical trial of 216 stable outpatients with HF, ejection fraction ≥50%, elevated *N*-terminal brain-type natriuretic peptide or elevated invasively measured filling pressures, and reduced exercise capacity. Participants were randomized from October 2008 through February 2012 at 26 centers in North America. Follow-up was through August 30, 2012.

Interventions Sildenafil (n=113) or placebo (n=103) administered orally at 20 mg, 3 times daily for 12 weeks, followed by 60 mg, 3 times daily for 12 weeks.

Main Outcome Measures Primary end point was change in peak oxygen consumption after 24 weeks of therapy. Secondary end points included change in 6-minute walk distance and a hierarchical composite clinical status score (range, 1-n, a higher value indicates better status; expected value with no treatment effect, 95) based on time to death, time to cardiovascular or cardiorenal hospitalization, and change in quality of life for participants without cardiovascular or cardiorenal hospitalization at 24 weeks.

Results Median age was 69 years, and 48% of patients were women. At baseline, median peak oxygen consumption (11.7 mL/kg/min) and 6-minute walk distance (308 m) were reduced. The median E/e' (16), left atrial volume index (44 mL/m^2), and pulmonary artery systolic pressure (41 mm Hg) were consistent with chronically elevated left ventricular filling pressures. At 24 weeks, median (IQR) changes in peak oxygen consumption (mL/kg/min) in patients who received placebo (−0.20 [IQR, −0.70 to 1.00]) or sildenafil (−0.20 [IQR, −1.70 to 1.11]) were not significantly different (*P*=.90) in analyses in which patients with missing week-24 data were excluded, and in sensitivity analysis based on intention to treat with multiple imputation for missing values (mean between-group difference, 0.01 mL/kg/min, [95% CI, −0.60 to 0.61]). The mean clinical status rank score was not significantly different at 24 weeks between placebo (95.8) and sildenafil (94.2) (*P*=.85). Changes in 6-minute walk distance at 24 weeks in patients who received placebo (15.0 m [IQR, −26.0 to 45.0]) or sildenafil (5.0 m [IQR, −37.0 to 55.0]; *P*=.92) were also not significantly different. Adverse events occurred in 78 placebo patients (76%) and 90 sildenafil patients (80%). Serious adverse events occurred in 16 placebo patients (16%) and 25 sildenafil patients (22%).

Conclusion and Relevance Among patients with HFPEF, phosphodiesterase-5 inhibition with administration of sildenafil for 24 weeks, compared with placebo, did not result in significant improvement in exercise capacity or clinical status.

Trial Registration clinicaltrials.gov Identifier: NCT00763867

JAMA. 2013;309(12):1268-1277
Published online March 11, 2013. doi:10.1001/jama.2013.2024 www.jama.com

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Heart failure with preserved ejection fraction (HFPEF) or diastolic heart failure is a common and highly morbid condition.[1] Clinical trials of renin-angiotensin system antagonists have not demonstrated improvement in outcomes or clinical status in HFPEF, and effective therapies are needed.[2] Phosphodiesterase-5 (PDE-5) metabolizes the nitric oxide and natriuretic peptide systems' second messenger cyclic guanosine monophosphate and thus may limit beneficial nitric oxide and natriuretic peptide actions in the heart, vasculature, and kidneys.

Preclinical studies suggest that inhibition of PDE-5 reverses adverse cardiac structural and functional remodeling and enhances vascular, neuroendocrine, and renal function.[3] In clinical studies, PDE-5 inhibitor therapy improved exercise tolerance and clinical status in patients with idiopathic pulmonary arterial hypertension and in patients with heart failure and reduced ejection fraction.[4-7] A small, single-center study of HFPEF observed improved hemodynamics, left ventricular diastolic function, right ventricular systolic function, left ventricular hypertrophy, and lung function with 6 to 12 months of therapy with a PDE-5 inhibitor compared with placebo.[8] In aggregate, these studies suggest the potential for PDE-5 inhibition to ameliorate several key pathophysiological perturbations in HFPEF, and thus improve exercise capacity and clinical status.

Accordingly, the Phosphodiesterase-5 Inhibition to Improve Clinical Status and Exercise Capacity in Heart Failure with Preserved Ejection Fraction (RELAX) trial was designed to test the hypothesis that, compared with placebo, therapy with the PDE-5 inhibitor sildenafil would improve exercise capacity in HFPEF after 24 weeks of therapy, assessed by the change in peak oxygen consumption.

METHODS

Study Oversight

All study participants provided written informed consent prior to screening. The National Heart, Lung, and Blood Institute–sponsored Heart Failure Clinical Research Network conceived, designed, and conducted the RELAX trial. The trial protocol was approved by a National Heart, Lung, and Blood Institute–appointed protocol review committee and data and safety monitoring board, and by the institutional review board at each participating site. The Duke Clinical Research Institute served as the data coordinating center.

Study Design

The rationale for, and design of, the RELAX trial have been described.[3] Patients with normal (≥50%) ejection fraction and heart failure with New York Heart Association functional class II through IV whose symptoms are stable while receiving medical therapy were eligible to participate if they had objective evidence of heart failure (previous heart failure hospitalization, or acute heart failure therapy with intravenous diuretic therapy, or chronic loop diuretic therapy for heart failure with left atrial enlargement, or invasively documented elevation in left ventricular filling pressures).

A peak oxygen consumption of 60% or less of the age- and sex–specific normal value[9] with a respiratory exchange ratio of 1.0 or more at the cardiopulmonary exercise test screening, and either elevated (≥400 pg/mL) *N*-terminal fragment of the precursor to brain-type natriuretic peptide (NT-proBNP) level or elevation in left ventricular filling pressures at the time of an NT-proBNP level less than 400 pg/mL were required for study entry. A complete list of the trial inclusion and exclusion criteria is provided in the online supplement (eTable 1, available at http://www.jama.com). As required in federally funded trials, a self-identification of investigator-defined race or ethnicity option was recorded.

Participants who met screening criteria underwent baseline studies (history and physical examination, cardiopulmonary exercise test, 6-minute walk distance, Minnesota Living with Heart Failure Questionnaire, echocardiography, cardiac magnetic resonance imaging [CMRI, if sinus rhythm], and phlebotomy for biomarkers), and were then randomly assigned, in a 1:1 ratio, to receive either sildenafil or placebo with the use of an automated web-based system. A permuted block randomization scheme was used with stratification according to clinical site and the presence of atrial fibrillation.

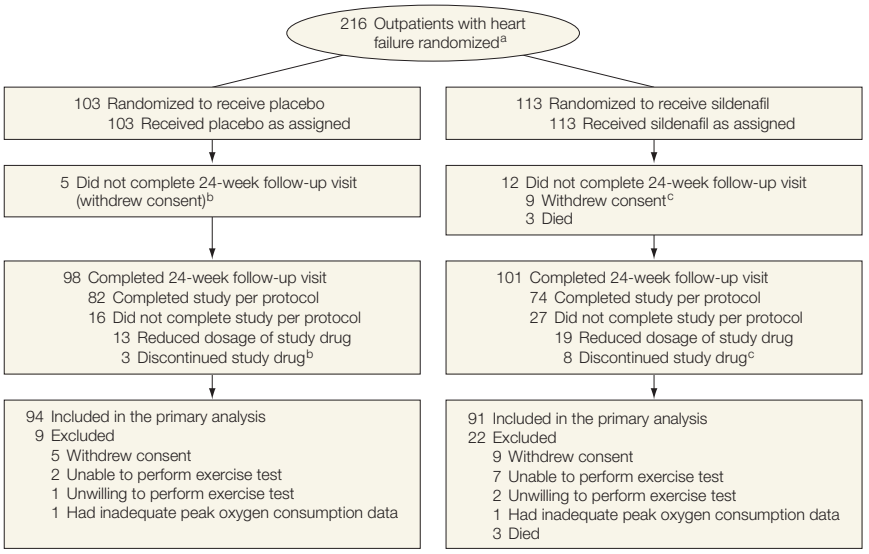
Study drug was administered orally at 20 mg, 3 times daily for 12 weeks. At the end of 12 weeks, each patient's history and physical examination, cardiopulmonary exercise test, 6-minute walk distance, and heart failure questionnaire were repeated. Phlebotomy for sildenafil levels was performed 2 hours after a scheduled dose of the study drug were obtained. The dose was then increased to 60 mg, 3 times daily for 12 weeks, after which baseline studies were repeated, including phlebotomy for sildenafil levels 2 hours after study drug. If adverse effects developed, study staff could recommend discontinuation or return to a lower or previously tolerated dose of the study drug.

Blinded core laboratories assessed biomarkers (University of Vermont), cardiopulmonary exercise tests (Massachusetts General Hospital, Harvard University), CMRIs (Duke University), and echocardiograms (Mayo Clinic).

Study End Points

The primary end point was the change in peak oxygen consumption after 24 weeks of therapy. A number of subgroup analyses were prespecified. Secondary end points included a composite hierarchical-rank clinical score in which patients were ranked (range, 1-n with data) based on time to death (tier 1), time to hospitalization for cardiovascular or cardiorenal causes (tier 2), and change in heart failure questionnaire from baseline (tier 3) for patients without cardiovascular or cardiorenal hospitalization after 24 weeks of therapy.[10] Because 189 patients had data for this end point, the anchor value (mean value in each group indicating no

Figure. Flow of Patients Through the Trial



Some patients discontinued study drug for a specific reason (>1 may apply) and later withdrew.
[a] Data regarding the number of patients screened for eligibility was not collected.
[b] Study drug discontinued due to adverse event (1), physician decision (1), other (2), or for withdrawal of consent (4).
[c] Study drug discontinued due to adverse event (9), physician decision (1), other (5), or for withdrawal of consent (5).

treatment effect) was 95. Other secondary end points included change in 6-minute walk distance at 24 weeks and change in peak oxygen consumption and 6-minute walk distance after 12 weeks of therapy. Peak sildenafil levels at 12 and 24 weeks and coinciding plasma cyclic guanosine monophosphate levels at 24 weeks were assessed. Using other prespecified end points, we assessed the effect of PDE-5 inhibition on left ventricular structure and vascular function by CMRI, Doppler-estimated diastolic function parameters and pulmonary artery systolic pressure, and biomarkers that reflect renal and neuroendocrine function, oxidative stress, and collagen metabolism.

The percent-predicted peak oxygen consumption, 6-minute walk distance, and the presence of chronotropic incompetence and left ventricular hypertrophy were calculated using published criteria (eMethods).[11-14]

Statistical Analysis

Power calculations were based on the standard deviation for change in peak oxygen consumption and the magnitude of its change when associated with improvements in other markers of clinical status (New York Heart Association class, 6-minute walk distance, and quality of life scores) in heart failure trials.[5,14,15] Based on these studies, a difference between treatment groups of 1.2 mL/kg/min in the change in peak oxygen consumption was considered clinically significant.

We estimated a 20% rate of incomplete primary end point data due to death, withdrawal, or incidence of new factors limiting ability to exercise. Using a 2-sample *t* test and a 2-sided α value of .05, a sample size of 190 patients had 85% power to detect a difference of 1.2 mL/kg/min in change in peak oxygen consumption, assuming 20% missing data and a standard deviation of change in peak oxygen consumption of 2.5 mL/kg/min. As an early-blinded interim analysis of aggregated primary end point completeness indicated that the missingness rate approached 20%, the blinded investigators recommended increasing the sample size to 215 patients.

The primary analyses were 2-sided, and patients without 24-week data were excluded. Sensitivity analysis for the primary end point was based on the intention to treat principle and utilized multiple imputations with 100 imputed data sets to account for missing 24-week data. In addition, a prespecified "last observation carried forward" sensitivity analysis used "carry-forward" of 12-week data if 24-week data were missing.

Data are presented as median (interquartile range [IQR]). For the comparison of treatment groups in the primary analysis, a multivariable linear-regression model was used adjusting for baseline peak oxygen consumption. A similar approach was used for the secondary end point of change in 6-minute walk distance, adjusting for baseline 6-minute walk distance. For the composite hierarchical-rank clinical score, patients were ranked independent of treatment assignment from 1 (worst outcome—the earliest death) to n (best outcome—survival with no cardiovascular or cardiorenal hospitalization, and the most favorable improvement in heart failure questionnaire) and treatments compared using the Wilcoxon rank sum test. For primary and secondary end points, a *P* value less than .05 was considered significant. For subgroup analyses, a treatment by subgroup interaction *P* less than .001 was considered significant.

All analyses were conducted with the use of SAS statistical software, version 9.2.

RESULTS

Patient Population

A total of 216 patients were enrolled in the trial from October 13, 2008, through February 21, 2012, at 26 sites in the United States and Canada (FIGURE). The baseline characteristics were not significantly different between treatment groups except for a lower prevalence of hypertension in the sildenafil group (TABLE 1). Patients in this study were older, 248% women, and obese with controlled blood pressure and multiple comorbidities including hypertension, ischemic heart disease, atrial fibrillation, diabetes, anemia, and chronic kidney disease.

Table 1. Baseline Characteristics of the Patients

	No. (%) of Patients		
	All (N = 216)	Placebo (n = 103)	Sildenafil (n = 113)
Age, median (IQR), y	69 (62-77)	69 (62-77)	68 (62-77)
Women	104 (48)	55 (53)	49 (43)
Self-reported white race	197 (91)	95 (92)	102 (90)
BMI, median (IQR)	32.9 (28.3-39.1)	32.8 (28.6-38.6)	33.3 (28.2-40.0)
NYHA functional classification			
II	101 (47)	46 (45)	55 (49)
III	115 (53)	57 (55)	58 (51)
MLHFQ total score, median (IQR)	43 (30-62)	43 (29-58)	44 (30-63)
Systolic blood pressure, median (IQR), mm Hg	126 (113-138)	127 (113-138)	124 (113-138)
Heart rate, median (IQR), beats/min	69 (61-78)	68 (62-77)	70 (61-80)
Jugular venous pressure ≥8 cm[a]	95/209 (45)	48/99 (48)	47/110 (43)
Edema	125 (58)	61 (59)	64 (57)
≥1 Heart failure hospitalizations in previous year	79 (37)	40 (39)	39 (35)
Qualifying ejection fraction, median (IQR), %	60 (55-66)	60 (55-65)	60 (55-66)
Medical history			
Hypertension	183 (85)	93 (90)	90 (80)
Ischemic heart disease	84 (39)	37 (36)	47 (42)
Atrial fibrillation or flutter	111 (51)	52 (50)	59 (52)
Diabetes mellitus	93 (43)	45 (44)	48 (42)
COPD	42 (19)	20 (19)	22 (19)
Anemia[b]	76 (35)	34 (33)	42 (38)
Local laboratory creatinine, median (IQR), mg/dL	1.2 (0.9-1.5)	1.1 (0.9-1.5)	1.3 (1.0-1.5)
Glomerular filtration rate, median (IQR), mL/min/1.73 m^2	57 (43-75)	59 (43-76)	57 (43-70)
Stage 3/4 chronic kidney disease	119 (55)	56 (54)	63 (56)
Medications at enrollment			
Loop diuretic	166 (77)	79 (77)	87 (77)
Any diuretic	186 (86)	86 (83)	100 (88)
ACE inhibitor or angiotensin II receptor blocker	152 (70)	78 (76)	74 (65)
β-Blocker	164 (76)	77 (75)	87 (77)
Aldosterone antagonist	23 (11)	9 (9)	14 (12)
Calcium channel blocker	66 (31)	35 (34)	31 (27)
Statin	138 (64)	67 (65)	71 (63)
Core laboratory biomarker data			
Cystatin C, median (IQR), mg/L[c]	1.31 (1.07-1.74)	1.34 (1.08-1.76)	1.29 (1.04,1.70)
NT-proBNP, median (IQR), pg/mL[d]	700 (283-1553)	648 (247-1679)	757 (318-1465)
Aldosterone, median (IQR), ng/dL[c]	18.9 (11.8-28.3)	18.0 (11.4-29.4)	20.7 (13.0-27.5)
Endothelin-1, median (IQR), pg/mL[c]	2.36 (1.95-3.20)	2.37 (2.03-3.14)	2.35 (1.85-3.22)
NT-procollagen III, median (IQR), μg/L[c]	7.7 (6.1-10.0)	7.9 (6.4-11.2)	7.5 (5.5-9.3)
Uric acid, median (IQR), mg/dL[e]	7.4 (5.8-8.5)	7.1 (5.8-8.5)	7.5 (5.8-8.5)
Peak functional status			
Oxygen consumption, median (IQR), mL/kg/min[f]	11.7 (10.2-14.4)	11.9 (10.1-14.4)	11.7 (10.4-14.5)
Predicted peak oxygen consumption, median (IQR), %	41 (35-49)	41 (36-49)	41 (34-48)
Respiratory exchange ratio, median (IQR)[f]	1.09 (1.02-1.15)	1.10 (1.03-1.15)	1.09 (1.02-1.16)
Systolic blood pressure, median (IQR), mm Hg[g]	156 (132-170)	154 (134-168)	156 (130-170)
Chronotropic incompetence[f]	164 (77)	79 (78)	85 (76)
6-Minute walk distance, median (IQR), m	308 (229-383)	305 (213-372)	308 (241-392)
Predicted 6-minute walk distance, median (IQR), %	69 (51-83)	68 (48-83)	70 (54-83)

Abbreviations: ACE, angiotensin-converting enzyme; BMI, body mass index, calculated as weight in kilograms divided by height in meters squared; COPD, chronic obstructive pulmonary disease; IQR, interquartile range; MLHFQ, Minnesota Living with Heart Failure Questionnaire; NT-proBNP, *N*-terminal fragment of the precursor to brain-type natriuretic peptide; NYHA, New York Heart Association.

SI conversion factors: To convert aldosterone from ng/dL to pmol/L, multiply by 27.74; creatinine from mg/dL to μmol/L, multiply by 88.4; and uric acid from mg/dL to μmol/L, multiply by 59.485.

[a] Indicates the number of patients/number of patients with nonmissing data for the variable (percentage).

[b] Anemia is defined as hemoglobin <13 g/dL for men, and <12 g/dL for women.

[c] Available in 214 (102 placebo and 112 sildenafil-treated) patients.

[d] Available in 213 (101 placebo and 112 sildenafil-treated) patients.

[e] Available in 212 (101 placebo and 111 sildenafil-treated) patients.

[f] Available in 215 (103 placebo and 112 sildenafil-treated) patients due to technical limitation precluding core laboratory measurement of peak oxygen consumption.

[g] Available in 209 (99 placebo and 110 sildenafil-treated) patients.

The heart failure questionnaire score was consistent with the distribution of New York Heart Association functional class observed in the study population. Evidence of volume overload (elevated jugular venous pressure or edema) and hospitalization for heart failure in the previous year were common. The majority of patients were taking diuretics, renin-angiotensin system antagonists, β-blockers, and statins. There was evidence of neuroendocrine activation and altered collagen metabolism as levels of NT-proBNP, aldosterone, endothelin-1, and NT-procollagen III exceeded reference ranges (eTable 2, available at http://www.jama.com). Both peak oxygen consumption and the 6-minute walk distance were reduced and chronotropic incompetence was common.

Baseline echocardiographic and CMRI characteristics of the patients were not significantly different between treatment groups (**Table 2**). At echocardiography, ejection fraction and left ventricular diastolic dimension (<5.6 cm) were within normal limits, while cardiac index was reduced. Nearly 50% of patients had left ventricular hypertrophy or evidence of concentric remodeling or hypertrophy (relative wall thickness ≥0.42).[16] There was Doppler evidence of diastolic dysfunction and elevated left ventricular filling pressures with reduced early diastolic medial mitral annular tissue velocity (e') and elevated ratio of early to late transmitral ventricular filling velocity (E/A), ratio of early transmitral filling velocity to early diastolic medial mitral annular tissue velocity (E/e'), left atrial volume, and pulmonary artery systolic pressure.

Overall, 132 patients (61%) were eligible for CMRI, and 117 of these underwent CMRI (89%). Ejection fraction and left ventricular end-diastolic volume index were within normal limits while cardiac index was reduced. In the CMRI cohort, 25% of patients had left ventricular hypertrophy. Aortic distensibility was reduced compared with published values in older participants without heart failure.[17]

Primary End Point

At 24 weeks, the median change in peak oxygen consumption from baseline was not significantly different in patients treated with placebo (−0.20 [IQR, −0.70 to 1.00]) and patients treated with sildenafil (−0.20 [IQR, −1.70 to 1.11]), (P=.90) (**Table 3**). Using multiple imputation to account for missing 24-week data, the mean difference between sildenafil and placebo was 0.01 mL/kg/min (favoring sildenafil, [95% CI, −0.60 to 0.61], P=.98). Carrying forward 12-week peak oxygen consumption when 24-week data were missing (placebo [n=5] and sildenafil [n=9]), median change in peak oxygen consumption (mL/kg/min) from baseline was −0.20 (IQR, −0.83 to 1.10) in patients treated with placebo and −0.13 (IQR, −1.50 to 1.16) in patients treated with sildenafil (P=.98). In subgroup analyses (eTable 3), the change in peak oxygen consumption was not significantly different between treatment groups when analysis was restricted to those patients still taking the study drug at week 24, in patients with or without left ventricular hypertrophy by CMRI, with lower or higher pulmonary artery systolic pressure, with lower or higher NT-

Table 2. Baseline Core Laboratory Echocardiographic and Cardiac Magnetic Resonance Imaging Characteristics of the Patients

	Placebo		Sildenafil		All Patients, Median (IQR) (N=216)
	No. of Patients (n = 103)	Median (IQR)	No. of Patients (n = 113)	Median (IQR)	
Doppler echocardiographic data					
Ejection fraction, %	103	60 (56-65)	110	60 (55-65)	60 (56-65)
Cardiac index, L/min/m^2	89	2.48 (2.06-2.86)	86	2.47 (2.09-2.92)	2.47 (2.07-2.92)
Left ventricular end-diastolic dimension, cm	76	4.6 (4.3-5.1)	88	4.6 (4.2-5.2)	4.6 (4.2-5.2)
Left ventricular mass/BSA, g/m^2	73	77 (63-90)	85	78 (61-97)	77 (62-96)
Left ventricular hypertrophy, No. (%)	73	36 (49)	85	40 (47)	76 (48)
Relative wall thickness ≥0.42, No. (%)	73	32 (44)	85	43 (51)	75 (48)
E/A ratio	70	1.3 (1.0-2.0)	72	1.6 (1.0-2.1)	1.5 (1.0-2.0)
Deceleration time, ms	95	190 (156-228)	98	182 (153-212)	185 (155-218)
Medial e', m/s	92	0.06 (0.05-0.07)	105	0.06 (0.05-0.08)	0.06 (0.05-0.08)
Medial E/e'	90	17 (12-24)	98	15 (10-23)	16 (11-23)
Left atrial volume index, mL/m^2	74	43 (38-59)	75	44 (33-59)	44 (35-59)
Pulmonary artery systolic pressure, mm Hg	75	41 (34-54)	63	41 (32-51)	41 (33-51)
CMRI data, median (IQR)					
Ejection fraction, %	56	66 (59-71)	61	66 (55-70)	66 (58-70)
Cardiac index, L/min/m^2	56	2.38 (1.94-2.80)	59	2.32 (2.11-2.74)	2.35 (1.98-2.76)
Left ventricular end-diastolic volume/BSA, mL/m^2	56	58 (49-68)	61	53 (46-66)	56 (47-67)
Left ventricular mass/BSA, g/m^2	56	61 (53-73)	61	65 (54-78)	63 (54-77)
Left ventricular hypertrophy, No. (%)	56	14 (25)	61	16 (26)	30 (26)
Aortic distensibility, median (IQR), 10^{-3} mm Hg^{-1}	41	1.08 (0.63-1.64)	45	1.18 (0.74-2.07)	1.17 (0.67-1.76)

Abbreviations: BSA, body surface area; CMRI, cardiac magnetic resonance imaging; e', early diastolic medial mitral annular tissue velocity; E/A, elevated ratio of early to late transmitral ventricular filling velocity; E/e', ratio of early transmitral filling velocity to early diastolic medial mitral annular tissue velocity; IQR, interquartile range.

proBNP, with or without atrial fibrillation or treated or not with renin-angiotensin system antagonists, β-blockers, or statins.

Secondary End Points and Safety Data

There were no significant differences in the clinical rank score, change in 6-minute walk distance at 24 weeks, or change in peak oxygen consumption or 6-minute walk distance at 12 weeks between treatment groups (Table 3). There were no significant differences in the components of the clinical rank score at 24 weeks or in the overall incidence of adverse, or serious adverse, events in the treatment groups.

Adverse events occurred in 78 patients (76%) who received placebo and 90 patients (80%) who received sildenafil. Serious adverse events occurred in 16 patients (16%) who received placebo and 25 patients (22%) who received sildenafil. Adverse events occurring in 5% or more of either study group are listed in eTable 4 (available at http://www.jama.com). Patients treated with sildenafil had a higher incidence of vascular adverse events, which included (but were not limited to) headache, flushing, and hypotension, although the change in mean arterial pressure from baseline to 24 weeks was not significantly different in patients treated with sildenafil (−1 [95% CI, −8 to 6]) and patients treated with placebo (−2 [95% CI, −10 to 7]), (P=.45). All serious adverse events exclusive of death or cardiovascular or cardiorenal hospitalization (Table 3) are listed in eTable 5. There were no other notable differences in the incidence of specific serious adverse events between study groups.

Study Drug and Cyclic Guanosine Monophosphate Levels

Median sildenafil concentrations measured approximately 2 hours after the last dose were 78 (IQR, 35-130 ng/mL) at 12 weeks and 200 (IQR, 92-330 ng/mL) at 24 weeks. At week 24, there was a weak correlation between sildenafil dose and sildenafil level (r=0.29, P=.008). In paired analysis, plasma cyclic guanosine monophosphate levels increased significantly from baseline to 24 weeks in patients randomized to receive sildenafil (mean increase, 8.72 pmol/mL, [95% CI, 2.56-14.87], P=.006), but not in patients randomized to placebo (mean increase, 1.28 pmol/mL, [95% CI, −6.27 to 8.83], P=.74), although the change in cyclic guanosine monophosphate was not significantly different between groups (P=.11).

Additional End Points

At CMRI, there was no difference in change in left ventricular mass or left ventricular end-diastolic volume index between treatment groups (TABLE 4). There was also no difference in change in Doppler-assessed left ventricular diastolic function parameters or pulmonary artery systolic pressure between treatment groups. By CMRI, arterial elastance decreased more and systemic vascular resistance tended to decrease more in patients treated with sildenafil (P=.09). However, the change in mean arterial pressure in the entire study population was not significantly different between groups, as noted above. More patients had missing data for aortic distensibility at 24 weeks than at baseline, but there was no difference in change in distensibility between groups. Patients treated with sildenafil had a greater increase in creatinine, cystatin C, NT-proBNP, uric acid, and endothelin-1 than patients treated with placebo, whereas changes in aldosterone and NT-procollagen III were not significantly different between groups.

COMMENT

To our knowledge, the RELAX trial is the first multicenter study to investigate the effect of PDE-5 inhibition in HFPEF. Contrary to our hypothesis, long-term PDE-5 inhibition in HFPEF had no effect on maximal or submaximal exercise capacity, clinical status, quality of life, left ventricular remod-

Table 3. Primary, Secondary, and Safety End Points

	Placebo		Sildenafil		
	No. of Patients	Variable	No. of Patients	Variable	*P* Value
Primary end point					
Change in peak oxygen consumption at 24 wk, median (IQR), mL/kg/min	94	−0.20 (−0.70 to 1.00)	91	−0.2 (−1.70 to 1.11)	.90
Secondary end points					
Clinical rank score, mean[a]	94	95.8	95	94.2	.85
Change in 6-minute walk distance at 24 wk, median (IQR), m	95	15.0 (−26.0 to 45.0)	90	5.0 (−37.0 to 55.0)	.92
Change in peak oxygen consumption at 12 wk, median (IQR), mL/kg/min	96	0.03 (−1.10 to 0.67)	97	0.01 (−1.35 to 1.25)	.98
Change in 6-minute walk distance at 12 wk, median (IQR), m	96	18.0 (−14.5 to 48.0)	99	10.0 (−25.0 to 36.0)	.13
Components of clinical rank score at 24 wk					
Death, No. (%)[b]	103	0	113	3 (3)	.25
Hospitalization for cardiovascular or renal cause, No. (%)	103	13 (13)	113	15 (13)	.89
Change in MLHFQ, median (IQR)	91	−8 (−21 to 5)	91	−8 (−19 to 0)	.44
Safety end points, No. (%)					
Adverse events	103	78 (76)	113	90 (80)	.49
Serious adverse events	103	16 (16)	113	25 (22)	.22

Abbreviations: IQR, interquartile range; MLHFQ, Minnesota Living with Heart Failure Questionnaire.
[a] A mean value of 95 in each group is expected under the null hypothesis of no treatment effect.
[b] Site investigator identified causes of death were sudden death (n=1), progressive cardiorenal failure (n=1), and noncardiovascular (n=1).

eling, diastolic function parameters, or pulmonary artery systolic pressure. Renal function worsened more and NT-proBNP, endothelin-1, and uric acid levels increased more in patients treated with sildenafil. Furthermore, there were more (but not significantly more) patients in the sildenafil group who withdrew consent, died, or were too ill to perform the cardiopulmonary exercise test, and patients treated with sildenafil had a higher incidence of vascular adverse events. These findings do not suggest that therapy with the PDE-5 inhibitor sildenafil provides clinical benefit in the general HFPEF population.

Given the strong rationale for testing PDE-5 inhibition in HFPEF[3] and the lack of benefit observed, it is important to consider whether the study population, specifics of the therapeutic intervention, and end points were appropriate.

The clinical characteristics of patients with HFPEF enrolled in major ongoing or completed clinical trials have been summarized.[18] The RELAX study population was similar to others in terms of age, sex distribution, and body size. Severity of heart failure in our study was also similar to or greater than other HFPEF trials, although comorbidity burden (diabetes, atrial fibrillation, kidney disease) may have been greater in this study population. Concentric remodeling and hypertrophy were common but not severe, and Doppler evidence of elevated filling pressures and pulmonary hypertension were present as was neuroendocrine activation consistent with the heart failure state.

However, the characteristics of the study population were notably different from the only other study to our knowledge that evaluated the effect of PDE-5 inhibition in HFPEF. In the study by Guazzi and coauthors,[8] sildenafil had a number of beneficial effects as outlined above, although effect on exercise capacity was not tested. Importantly, in that study, patients with HFPEF had fewer comorbidities and significantly higher blood pressure, left ventricular mass, and pulmonary artery systolic pressure than the patients in the RELAX study and catheterization-documented pulmonary arterial hypertension, profound right ventricular systolic dysfunction, and right ventricular failure were present. This profile is somewhat atypical for HFPEF cohorts.[19]

It may be that the primary therapeutic effects of PDE-5 inhibitors in heart failure involve the drugs' ability to dilate the pulmonary vascular bed, enhance right ventricular contractility, and reduce ventricular interdependence,[4,19-22] and that pulmonary arterial hypertension and right ventricular failure must be significant in order to observe clinical benefit in HFPEF. Our subgroup analysis did not show any trends toward improvement in peak oxygen consumption in patients with higher pulmonary artery systolic pressure, but the presence of pulmonary arterial hypertension or right ventricular dysfunction was not assessed in this study.

Although left ventricular hypertrophy was common in participants in this study, it was far less severe than among participants in the study by Guazzi et al.[8] In murine-pressure overload studies, PDE-5 inhibition did not have an-

Table 4. Additional Prespecified End Points

	Placebo		Sildenafil		
	No. of Patients	Median (IQR)	No. of Patients	Median (IQR)	*P* Value
Change in left ventricular structure by CMRI at 24 wk					
Left ventricular mass by CMRI, g	47	0.6 (−5.7 to 7.9)	49	−1.5 (−5.9 to 7.1)	.93
Left ventricular end-diastolic volume by CMRI, mL	47	−4.3 (−15.5 to 8.1)	49	3.7 (−4.9 to 14.5)	.13
Change in diastolic function parameters at 24 wk					
Medial e', m/s	83	0.00 (−0.01 to 0.01)	77	0.00 (−0.01 to 0.01)	.88
E/e'	80	−1.6 (−4.7 to 2.2)	75	0.2 (−2.4 to 3.1)	.16
PA systolic pressure, mm Hg	58	−2 (−8 to 8)	45	2 (−5 to 7)	.94
Change in vascular function by CMRI at 24 wk					
Arterial elastance, mm Hg/mL	45	0.03 (−0.14 to 0.23)	47	−0.08 (−0.41 to 0.12)	.02
Systemic vascular resistance, wood units	45	0.13 (−0.15 to 0.43)	47	−0.04 (−0.38 to 0.21)	.09
Aortic distensibility, 10^{-3} mm Hg^{-1}	31	0.13 (−0.17 to 0.42)	29	0.18 (−0.12 to 0.67)	.38
Change in core laboratory biomarkers at 24 wk					
Creatinine, mg/dL	94	0.01 (−0.10 to 0.09)	94	0.05 (−0.04 to 0.15)	.047
Cystatin C, mg/L	95	0.01 (−0.08 to 0.11)	95	0.05 (−0.04 to 0.16)	.01
NT-proBNP, pg/mL	94	−23 (−198 to 139)	95	15 (−90 to 372)	.03
Endothelin-1, pg/mL	95	−0.01 (−0.48 to 0.47)	95	0.38 (−0.10 to 0.97)	.046
Aldosterone, ng/dL	95	0 (−7.0 to 4.8)	95	−1.1 (−7.7 to 3.0)	.85
NT-procollagen III, μg/L	93	−0.03 (−1.49 to 1.54)	95	0.07 (−1.17 to 1.42)	.77
Uric acid, mg/dL	94	−0.1 (−0.7 to 0.7)	94	0.3 (−0.4 to 1.4)	.02

Abbreviations: CMRI, cardiac magnetic resonance imaging; e', early diastolic medial mitral annular tissue velocity; E/e', ratio of early transmitral filling velocity to early diastolic medial mitral annular tissue velocity; IQR, interquartile range; NT-proBNP, *N*-terminal fragment of the precursor to brain-type natriuretic peptide; PA, pulmonary artery.
SI conversions: To convert aldosterone from ng/dL to pmol/L, multiply by 27.74; creatinine from mg/dL to μmol/L, multiply by 88.4; and uric acid from mg/dL to μmol/L, multiply by 59.485.

tihypertrophic effects in mice with less severe pressure overload and compensated left ventricular hypertrophy with relatively preserved ejection fraction, whereas dramatic antiremodeling benefits were observed in mice with severe pressure overload, eccentric left ventricular hypertrophy, reduced ejection fraction, and pulmonary congestion.[23,24] Conceivably, activation of PDE-5 or of cyclic guanosine monophosphate–sensitive downstream pathways in the left ventricle or other organs may occur only in heart failure associated with advanced left ventricular remodeling.

In a randomized clinical trial of PDE-5 inhibition in pulmonary arterial hypertension, the effect of sildenafil on exercise capacity was not dose related as improvement in 6-minute walk distance was seen with 20 mg 3 times daily after just 4 weeks of therapy with no further improvement with higher doses or longer duration of therapy.[4] Sildenafil levels at 12 and 24 weeks were variable, but on average, similar to random levels observed with similar doses among participants with heart failure and reduced ejection fraction for whom improvement in exercise capacity with long-term sildenafil use was observed (Gregory Lewis, MD, Massachusetts General Hospital, written communication, February 2, 2013).[5]

For patients with heart failure and reduced ejection fraction, the benefit of PDE-5 inhibition on exercise capacity has been demonstrated acutely after a single 50-mg dose,[25,26] and with 75 mg, 3 times daily for 12 weeks (dose uptitrated over the course of 6 weeks).[5] Although only 73% of patients in the sildenafil group attending the 24-week follow-up were taking the per-protocol dose, 92% of patients were taking at least 20 mg 3 times daily. While studies in pulmonary arterial hypertension and heart failure and reduced ejection fraction have observed effects on exercise capacity with similar doses and duration of therapy, we cannot exclude the possibility that inadequate dose or duration of PDE-5 inhibition contributed to our findings.

Therapeutic sildenafil levels were associated with minimal increases in plasma cyclic guanosine monophosphate. Heart failure with preserved ejection fraction is characterized by endothelial dysfunction[27] and by lower natriuretic peptide levels than observed in heart failure and reduced ejection fraction,[28] which may suggest limited nitric oxide and natriuretic peptide activity in HFPEF. Inability to enhance cyclic guanosine monophosphate with PDE-5 inhibition in HFPEF may have contributed to our findings.

As previously described,[3] change in peak oxygen consumption was chosen as the primary end point in the RELAX trial based on previous preclinical and clinical studies and because noncardiovascular comorbidities and motivational factors can influence measures of submaximal exercise performance in HFPEF. Our trial was powered to detect a clinically significant difference in the change in peak oxygen consumption between groups and the estimate of variability (SD, 2.5 mL/kg/min) in change in peak oxygen consumption used in the power calculations was consistent with the standard deviation of change in peak oxygen consumption observed in the patients treated with placebo (2.0 mL/kg/min). The lack of treatment effect on submaximal exercise, clinical status, and physiologic end points supports the validity of the observed lack of treatment effect on maximal exercise capacity.

The high prevalence of chronotropic incompetence in the study population is noteworthy. Chronotropic incompetence may contribute to exercise limitation in HFPEF, and may not be improved by PDE-5 inhibition.

Although numerous studies in animal models of renal dysfunction suggest that PDE-5 inhibition ameliorates progression of renal dysfunction of various etiologies,[3,29-31] in this trial, modest but statistically significant worsening of renal function was observed in patients treated with sildenafil and was associated with concordant increases in NT-proBNP, uric acid, and endothelin-1, suggesting that the decline in renal function was physiologically significant. Studies in pulmonary arterial hypertension and erectile dysfunction have not reported worsening of renal function with PDE-5 inhibitor therapy, but little is known of the effect of PDE-5 inhibition on renal function in HFPEF.

There were more (but not significantly more) patients who withdrew consent, died, or were too ill to perform the cardiopulmonary exercise test in the sildenafil treatment group, potentially accentuating the lack of benefit observed, particularly if those who withdrew did so due to adverse effects or poor clinical status.

A modest decrease in arterial elastance was noted in patients treated with sildenafil in the CMRI cohort. This may have been related to an effect of sildenafil on resistance that tended to decrease more in patients treated with sildenafil, but in the entire cohort, there were no differences in change in mean arterial pressure between treatment groups.

The findings of this study must be interpreted in the context of other potential limitations. Multicenter trials using peak oxygen consumption as a primary end point are challenging, but rigorous methodologies were used in the design and execution of the exercise test study protocol.[3] Patients were selected who could perform the test, and who had significant reduction in peak oxygen consumption—these entry criteria may have selected for a unique HFPEF phenotype. The trial was not powered to address differences in clinical outcomes.

CONCLUSIONS

Among patients with HFPEF, PDE-5 inhibition with administration of sildenafil for 24 weeks, compared with placebo, did not result in significant improvement in exercise capacity or clinical status. Continued efforts to identify key pathophysiologic perturbations and novel therapeutic targets in HFPEF are needed.

Published Online: March 11, 2013. doi:10.1001/jama.2013.2024

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For a complete listing of the RELAX site representatives see the online supplement at jama.com.

Author Contributions: Drs Lee and Anstrom had full access to all the data in the study and take responsibility for the integrity of the data and the accuracy of the data analysis.
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Obtained funding: Chen, Semigran, Lee, Bull, Deswal, Ofili, Felker, McNulty, Velazquez, Hernandez, Mascette, Redfield.
Administrative, technical, or material support: Chen, Borlaug, Lee, Rouleau, Bull, O'Connor, McNulty, Ibarra, Oh, Kim, Tracy, Velazquez, Hernandez, Mascette, Braunwald.
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Conflict of Interest Disclosures: All authors have completed and submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Dr Redfield reported receiving financial support from the National Institutes of Health (NIH) and royalties from Annexon. Dr Chen reported receiving funding for patents that his institution has licensed to Nile Therapeutics and Annexon with other patents pending at the US Patent and Trademark Office. He also reported that he has royalties with Nile Therapeutics, Annexon, and Up To Date. Dr Borlaug reported receiving consulting fees from Medscape, Amgen, and GlaxoSmithKline and receiving financial support from Atcor Medical and Gilead. Dr Semigran reported receiving financial support from the NIH for participation in the Heart Failure Network and from Bayer. Dr Lee reported receiving financial support from the NIH for participation in the Heart Failure Network. Dr LeWinter reported receiving financial support from the NIH for participation in the Heart Failure Network. Dr Bull reported receiving financial support from the NIH for participation in the Heart Failure Network. Dr Deswal reported receiving financial support from the NIH for participation in the Heart Failure Network. Dr Stevenson reported receiving financial support from the NIH for participation in the Heart Failure Network. Dr Givertz reported receiving financial support from the NIH for participation in the Heart Failure Network. Dr Ofili reported receiving financial support from the NIH, receiving consulting fees from Merck, Novartis, and Arbor Pharmaceuticals and lecture fees from Arbor Pharmaceuticals. She is currently employed by the Morehouse School of Medicine and has grants pending from the National Heart, Lung, and Blood Institute and National Institute on Minority Health and Health Disparities. Dr O'Connor reported receiving consulting fees from Roche Diagnostics, Amgen, Novartis, Ikaria, Acetlion Pharma, Heartware, ResMed, Pozen, GE Healthcare, Johnson & Johnson, Gilead, Critical Diagnostics, BG Medicine, Otsuka, Astellas, Novella, Cytokinetics, and Capricor; holding stock options with Neurotronik/Interventional Autonomics Corporation; editing *American College of Cardiology*; and coowning Cardiology Consulting Associates. Dr Felker reported receiving financial support from the NIH; consulting fees from Novartis, Amgen, Trevena, Roche Diagnostics, Merck, Singulex, and BG Medicine; and funding from Amgen, Otsuka, and Roche Diagnostics. Dr Bart reported receiving financial support from the NIH for participation in the Heart Failure Network. Ms Ibarra reported receiving financial support from the NIH for participation in the Heart Failure Network. Dr Oh received financial support from the NIH for the Echo Core Lab. Dr Patel has received consulting fees from Genzyme, Baxter, Jensen, and Bayer, and his financial institution receives financial support from AstraZeneca, Johnson & Johnson, and Maquet. Dr Kim reported receiving financial support from the NIH as the Heart Failure Network Data Coordinating Center and MRI Core Lab, and funding for a patent with Northwestern University. His institution has received a grant from Siemens. Dr Tracy reported receiving financial support from the NIH for participation in the Heart Failure Network, and consulting fees from Merck, Tibotec—Johnson & Johnson, and Abbott. He has also given expert testimony about biomarkers in blood clotting and is the owner of Haematologic Technologies. Dr Velazquez reported receiving financial support from the NIH for participation in the Heart Failure Network. Dr Anstrom reported receiving financial support from the NIH for participation in the Heart Failure Network. Dr Hernandez reported receiving financial support from the NIH for participation in the Heart Failure Network and consulting fees from AstraZeneca, Corthera, Janseen, and Bristol-Myers Squibb. His institution has received funding from Amylin, Johnson & Johnson, and Bristol-Myers Squibb. Dr Braunwald reported receiving consulting fees from Merck, Daiichi Sankyo, Genzyme, Amorcyte, Medicines Co, MC Communications, Ikaria, CardioRentis, sanofi-aventis; funding (or pending funding) from AstraZeneca, Johnson & Johnson, Merck, sanofi-aventis, Daiichi Sankyo, GlaxoSmithKline, Bristol-Myers Squibb, Beckman Coulter, Roche Diagnostics, and Pfizer; compensation for lectures for Eli Lilly, Merck, CVRx (no compensation), CV Therapeutics (now Gilead), Daiichi Sankyo, MC Communications, Manarini International, Medscape, and Bayer; and payment for development of educational presentations from MC Communications. No other disclosures were reported.

Funding/Support: This work was supported by grants U10HL084904 (for the data coordinating center), and U10HL084861, U10HL084875, U10HL084877, U10HL084889, U10HL084890, U10HL084891, U10HL084899, U10HL084907, and U10HL084931 (for the clinical centers) from the National Heart, Lung, and Blood Institute. This work is also supported by grant UL1TR000454 from the National Center for Advancing Translational Sciences and grant 8 U54 MD007588 from the National Institute on Minority Health and Health Disparities. Pfizer provided study drug (sildenafil and matched placebo).
Role of the Sponsor: Pfizer had no role in the design and conduct of the study, collection, management, analysis, or interpretation of the data, or preparation, review, or approval of the manuscript. National Heart, Lung, and Blood Institute representatives provided advice on the trial design and appointed the protocol review committee and data and safety monitoring board.
Disclaimer: The authors are solely responsible for the content of this article, which does not necessarily represent the official views of the National Heart, Lung, and Blood Institute or the National Institutes of Health.
Online-Only Material: The eMethods, eReferences, eTables 1 through 5, and eAppendices are available at http://www.jama.com.
Additional Contributions: We thank the patients who participated in this study, the Heart Failure Network site investigators and coordinators, the members of the Heart Failure Network data and safety monitoring board and protocol review committee, and the National Heart, Lung, and Blood Institute representatives. For a complete listing of the Heart Failure Network members, see Appendix C available at http://www.jama.com.

REFERENCES

1. Owan TE, Redfield MM. Epidemiology of diastolic heart failure. *Prog Cardiovasc Dis*. 2005;47(5):320-332.
2. Shah RV, Desai AS, Givertz MM. The effect of renin-angiotensin system inhibitors on mortality and heart failure hospitalization in patients with heart failure and preserved ejection fraction: a systematic review and meta-analysis. *J Card Fail*. 2010;16(3):260-267.
3. Redfield MM, Borlaug BA, Lewis GD, et al; Heart Failure Clinical Research Network. PhosphdiesteRasE-5 inhibition to improve clinical status and exercise capacity in diastolic heart failure (RELAX) trial: rationale and design. *Circ Heart Fail*. 2012;5(5):653-659.
4. Galiè N, Ghofrani HA, Torbicki A, et al; Sildenafil Use in Pulmonary Arterial Hypertension (SUPER) Study Group. Sildenafil citrate therapy for pulmonary arterial hypertension. *N Engl J Med*. 2005;353(20):2148-2157.
5. Lewis GD, Shah R, Shahzad K, et al. Sildenafil improves exercise capacity and quality of life in patients with systolic heart failure and secondary pulmonary hypertension. *Circulation*. 2007;116(14):1555-1562.
6. Guazzi M, Vicenzi M, Arena R. Phosphodiesterase 5 inhibition with sildenafil reverses exercise oscillatory breathing in chronic heart failure: a long-term cardiopulmonary exercise testing placebo-controlled study. *Eur J Heart Fail*. 2012;14(1):82-90.
7. Guazzi M, Vicenzi M, Arena R, Guazzi MD. PDE5 inhibition with sildenafil improves left ventricular diastolic function, cardiac geometry, and clinical status in patients with stable systolic heart failure: results of a 1-year, prospective, randomized, placebo-controlled study. *Circ Heart Fail*. 2011;4(1):8-17.

8. Guazzi M, Vicenzi M, Arena R, Guazzi MD. Pulmonary hypertension in heart failure with preserved ejection fraction: a target of phosphodiesterase-5 inhibition in a 1-year study. *Circulation*. 2011;124(2):164-174.
9. Fletcher GF, Balady G, Froelicher VF, Hartley LH, Haskell WL, Pollock ML; Writing Group. Exercise standards: a statement for healthcare professionals from the American Heart Association. *Circulation*. 1995;91(2):580-615.
10. Packer M. Proposal for a new clinical end point to evaluate the efficacy of drugs and devices in the treatment of chronic heart failure. *J Card Fail*. 2001;7(2):176-182.
11. Enright PL, Sherrill DL. Reference equations for the six-minute walk in healthy adults. *Am J Respir Crit Care Med*. 1998;158(5 Pt 1):1384-1387.
12. Khan MN, Pothier CE, Lauer MS. Chronotropic incompetence as a predictor of death among patients with normal electrograms taking beta blockers (metoprolol or atenolol). *Am J Cardiol*. 2005;96(9):1328-1333.
13. Chirinos JA, Segers P, De Buyzere ML, et al. Left ventricular mass: allometric scaling, normative values, effect of obesity, and prognostic performance. *Hypertension*. 2010;56(1):91-98.
14. Feldman AM, Silver MA, Francis GS, et al; PEECH Investigators. Enhanced external counterpulsation improves exercise tolerance in patients with chronic heart failure. *J Am Coll Cardiol*. 2006;48(6):1198-1205.
15. Abraham WT, Fisher WG, Smith AL, et al; MIRACLE Study Group, Multicenter InSync Randomized Clinical Evaluation. Cardiac resynchronization in chronic heart failure. *N Engl J Med*. 2002;346(24):1845-1853.
16. Lang RM, Bierig M, Devereux RB, et al; Chamber Quantification Writing Group; American Society of Echocardiography's Guidelines and Standards Committee; European Association of Echocardiography. Recommendations for chamber quantification: a report from the American Society of Echocardiography's Guidelines and Standards Committee and the Chamber Quantification Writing Group, developed in conjunction with the European Association of Echocardiography, a branch of the European Society of Cardiology. *J Am Soc Echocardiogr*. 2005;18(12):1440-1463.
17. Hundley WG, Kitzman DW, Morgan TM, et al. Cardiac cycle-dependent changes in aortic area and distensibility are reduced in older patients with isolated diastolic heart failure and correlate with exercise intolerance. *J Am Coll Cardiol*. 2001;38(3):796-802.
18. Shah SJ, Heitner JF, Sweitzer NK, et al. Baseline characteristics of patients in the Treatment of Preserved Cardiac Function Heart Failure with an Aldosterone Antagonist (TOPCAT) trial. *Circ Heart Fail*. 2012.
19. Forfia PR, Borlaug BA. Letter by Forfia and Borlaug regarding article, "Pulmonary hypertension in heart failure with preserved ejection fraction: a target of phosphodiesterase-5 inhibition in a 1-year study." *Circulation*. 2012;125(8):e408-e410. doi:10.1161/CIRCULATIONAHA.111.064584.
20. Nagendran J, Archer SL, Soliman D, et al. Phosphodiesterase type 5 is highly expressed in the hypertrophied human right ventricle, and acute inhibition of phosphodiesterase type 5 improves contractility. *Circulation*. 2007;116(3):238-248.
21. Xie YP, Chen B, Sanders P, et al. Sildenafil prevents and reverses transverse-tubule remodeling and Ca(2+) handling dysfunction in right ventricle failure induced by pulmonary artery hypertension. *Hypertension*. 2012;59(2):355-362.
22. Borgdorff MA, Bartelds B, Dickinson MG, et al. Sildenafil enhances systolic adaptation, but does not prevent diastolic dysfunction, in the pressure-loaded right ventricle. *Eur J Heart Fail*. 2012;14(9):1067-1074.
23. Nagayama T, Hsu S, Zhang M, et al. Pressure-overload magnitude-dependence of the anti-hypertrophic efficacy of PDE5A inhibition. *J Mol Cell Cardiol*. 2009;46(4):560-567.
24. Takimoto E, Champion HC, Li M, et al. Chronic inhibition of cyclic GMP phosphodiesterase 5A prevents and reverses cardiac hypertrophy. *Nat Med*. 2005;11(2):214-222.
25. Lewis GD, Lachmann J, Camuso J, et al. Sildenafil improves exercise hemodynamics and oxygen uptake in patients with systolic heart failure. *Circulation*. 2007;115(1):59-66.
26. Guazzi M, Tumminello G, Di Marco F, Fiorentini C, Guazzi MD. The effects of phosphodiesterase-5 inhibition with sildenafil on pulmonary hemodynamics and diffusion capacity, exercise ventilatory efficiency, and oxygen uptake kinetics in chronic heart failure. *J Am Coll Cardiol*. 2004;44(12):2339-2348.
27. Borlaug BA, Olson TP, Lam CS, et al. Global cardiovascular reserve dysfunction in heart failure with preserved ejection fraction. *J Am Coll Cardiol*. 2010;56(11):845-854.
28. Bishu K, Deswal A, Chen HH, et al. Biomarkers in acutely decompensated heart failure with preserved or reduced ejection fraction. *Am Heart J*. 2012;164(5):763-770 e763.
29. Tapia E, Sanchez-Lozada LG, Soto V, et al. Sildenafil treatment prevents glomerular hypertension and hyperfiltration in rats with renal ablation. *Kidney Blood Press Res*. 2012;35(4):273-280.
30. Medeiros PJ, Villarim Neto A, Lima FP, Azevedo IM, Leão LR, Medeiros AC. Effect of sildenafil in renal ischemia/reperfusion injury in rats. *Acta Cir Bras*. 2010;25(6):490-495.
31. Choi DE, Jeong JY, Lim BJ, et al. Pretreatment of sildenafil attenuates ischemia-reperfusion renal injury in rats. *Am J Physiol Renal Physiol*. 2009;297(2):F362-F370.