Supplementary Appendix

This appendix has been provided by the authors to give readers additional information about their work.

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ONLINE SUPPLEMENT

Table of Contents

Section 1: Methods - Endotrophin assay

The method for measurement, technical specifications and details of the PRO-C6 assay have been described previously by Sun et al.1 The PRO-C6 assay is produced under a regulated environment (good manufacturing practice (GMP)) following strict quality standards. The assay is constantly evaluated for variability and reproducibility by analyzing lot-to-lot controls and assessed in relation to critical assay parameters, including variance in the standard curve and lower- and upper limits of detection. Technical results on the reproducibility of the PRO-C6 assay are shown in **Table S2**. Reproducibility was measured by assessing the recovery of the analyte spiked in a matrix. This was done to ensure that no factors present in the sample matrix will interfere with the signal obtained by the binding between antibody and analyte. Ideally, the sample recovery should be between 80-120%. As shown in **Table S2**, the PRO-C6 assay recovery varied from 96.0% to 100.1% (mean 99.2%, SD 1.9%), well within the technical limit for an adequate signal-to-noise ratio. The inter- and intra assay variations are also reported over 6 different lots. Inter- and intra assay variability are used to express the precision and repeatability of the test results. Inter-assay variability is a measure of variance between sample values measured on different assay plates and is thus reflective of plate-to-plate variance. Intra-assay variability is a measure of variance between sample values run on the same plate, and variance The PRO-C6 assay demonstrated a mean inter-assay variance of 11.9% (SD 3.1%), and an intra-assay variance of 5.2% (SD 1.9%). Also reported in **Table S2** is the starting point (Std A) for the respective PRO-C6 standard curves over 6 lots. The mean value was 108.9 ng/ml (SD 9.7 ng/ml). The values for two internal kit controls (1 and 2) are also reported, with mean values of 4.8 ng/ml (SD 0.5 ng/ml) and 14.6 ng/ml (SD 1.3 ng/ml). The PRO-C6 standard curve slope is also shown, as measured in the linear area of the curve. Optimally, the value should be close to

1.0, which means that a change in concentration of the analyte will lead to an equal relative change in signal measured by optical density (OD). The standard curve slope for the PRO-C6 assay exhibited a mean of 0.9 (SD 0.03) over 6 different lots. The IC50 value of the standard curve is also reported, which is a measure of the sensitivity of the binding between antibody and analyte. Generally, the lower the IC50, the more sensitive the assay will be. Considering the standard curve of the PRO-C6 assay, we can conclude that the PRO-C6 assay exhibited high sensitivity for detecting the analyte (mean IC50 of 4.8 ng/ml, SD 1.0%).

Long-term stability of endotrophin in frozen samples was studied 3 human samples spanning storage of 3 years at -70 °C. The signal recovery was assessed in relation to the levels obtained from samples stored at -150 °C for an equal period of time. When stored at -70 °C, mean recovery at 1, 2 and 3 years was 93.8%, 92.2% and 93.9%, respectively. These data suggest adequate analyte stability up to at least 3 years.

Section 1: Methods - Study populations

Aim 1: TOPCAT trial

Inclusion criteria for TOPCAT were as follows: age ≥50 years; diagnosis of HF based on at least 1 HF symptom at the time of study screening, and at least 1 HF sign within the 12 months before screening; left ventricular EF ≥45%; at least 1 HF hospitalization in the 12 months before study screening or BNP (B-type natriuretic peptide) >100 pg/mL or NT-proBNP (N-terminal pro-BNP) >360 pg/mL (in the absence of an alternative explanation for elevated natriuretic peptide level) within the 60 days before screening; and serum potassium <5.0 mmol/L before randomization.

Exclusion criteria have been published in detail previously but included severe systemic illness with a life expectancy of <3 years, significant chronic pulmonary disease, infiltrative or hypertrophic cardiomyopathy, constrictive pericarditis, previous cardiac transplant or left ventricular assist device, known chronic hepatic disease, severe chronic kidney disease (defined as estimated glomerular filtration rate [eGFR] <30 mL/min per 1.73 m^2 or serum creatinine ≥2.5 mg/dL), a history of significant hyperkalemia, known intolerance to aldosterone antagonists, and recent myocardial infarction, coronary artery bypass grafting, or percutaneous coronary intervention.

Samples were obtained for 206 participants who were enrolled in the Americas. Due to volume availability, measurements were completed in 205 participants. Given that natriuretic peptide data in the parent trial were available only in a minority of participants enrolled from the Americas, we performed de novo NT-proBNP measurements in frozen plasma samples a standardized manner using a validated Luminex assay (Bristol-Myers Squibb, Ewing Township, New Jersey), blinded to participant clinical data and outcomes.

Aim 2: External validation of the relationship between endotrophin and outcomes in HFpEF

We validated the relationships between endotrophin and outcomes in additional cohort studies in which HFpEF participants were prospectively identified, enrolled and followed for adjudicated outcomes (Table 1). We included HFpEF participants from the Penn Heart Failure Study (USA; n=174),[2-5] the PEOPLE cohort (New Zealand; n=168)[6], a randomized trial of vasodilator therapy in HFpEF (n=45),[7] a HFpEF cohort from University of Pamplona, Spain (n=171) and a HFpEF cohort from University of Valencia, Spain (n=47). We performed a participant-level meta-

analysis of all available cohorts (n=810) to assess the relationship between endotrophin and 1) All cause-death; 2) DHFA.

Penn Heart Failure Study: The PHFS design has been previously published.[2-5] Briefly, the PHFS was a prospective cohort study of ambulatory patients with chronic HF recruited between 2003-2011 at the University of Pennsylvania (Philadelphia, PA), Case Western Reserve University (Cleveland, OH), and the University of Wisconsin (Madison, WI). Patients with a clinical diagnosis of HF as determined by a HF specialist were enrolled. Each participant provided written informed consent. At the time of study entry, standardized questionnaires were administered to participants and their physicians to obtain detailed clinical data. Participants with expected mortality of 6 months or less from a non-cardiac condition, mechanical circulatory support, or inability to provide informed consent were excluded. Venous blood samples were obtained at enrollment and stored at -80 °C for later analysis. An institutional review board from each participating center approved the protocol. All participants with HFpEF with available plasma samples were included in this analysis (n=174).

PEOPLE cohort: The Prospective Evaluation of Outcome in Patients with Heart Failure with Preserved Left Ventricular Ejection Fraction (PEOPLE) was a prospective longitudinal study of adults with HF from four New Zealand centres.[6,8] All centres were university or public hospitals serving large proportions of the community. Patients were those over age 18 years with a clinical diagnosis of HF, according to the attending physician and verified by the site investigator according to 2012 European Society of Cardiology criteria. Recruitment occurred either when the patient was in hospital (70%) for a primary diagnosis of HF (assessment was done following

stabilization of the acute HF) or in the outpatient setting (30%) within 6 months of an episode of decompensated HF (requiring hospitalization or treatment in an out-patient setting). Exclusion criteria included severe valve disease, transient acute pulmonary oedema in the context of primary acute coronary syndrome, end-stage renal failure, specific HF subgroups (including constrictive pericarditis, congenital heart disease, hypertrophic cardiomyopathy, cardiac amyloid, and chemotherapy-associated cardiomyopathy), isolated right HF, life-threatening co-morbidity with life expectancy <1 year and inability to provide consent.

Background demographic characteristics and clinical history were extensively documented. Echocardiography was undertaken following a standardized protocol (American Society of Echocardiography guidelines). Heart failure with preserved ejection fraction was defined as LVEF >50%. Systematic follow-up captured clinical events including all-cause mortality, recurrent decompensated heart failure and other cardiovascular adverse events from minimum of 2 years from recruitment. Of 4789 patients potentially eligible, 869 were unable to provide informed consent, and a further 2979 met one or more of the study exclusion criteria, leaving 941 patients included in the study. Of these 331 had HFpEF including the 168 participants contributing data to the current analysis. Endotrophin was measured in plasma.

Leizarán Cohort: The details of this cohort have been previously published.[9,10] Leizarán is a prospective cohort study of ambulatory hypertensive patients (systolic blood pressure and diastolic blood pressure of >139 and/or 89 mmHg, respectively, or under antihypertensive treatment) with chronic HF recruited between 2002 and 2010 at the Hospital Universitario Donostia (Spain). All of participants gave written informed consent, and the institutional review committee approved the study protocol. All patients had a previous clinical diagnosis of chronic

heart failure (HF) based on the presence of at least one major and two minor Framingham criteria. All patients had presented previously at least one hospitalization for HF. All patients were in New York Heart Association (NYHA) functional classes II to IV. All participants with HFpEF with available serum samples were included in this analysis (n=171). Patients were followed for a median of 5.31 years (range, 0.24–7.21 years). Endotrophin was measured in serum.

University of Valencia study (TRAINING-HF trial): It was a randomized clinical trial performed in the Hospital Clínico Valencia-University of Valencia in which we evaluated the effect of inspiratory muscle training, functional electrical stimulation or a combination of both on peak exercise oxygen uptake at 12 and 24-week in patients with stable symptomatic patients (New York Heart Association II-III) with HFpEF. The eligibility of candidate patients was based on the following inclusion criteria: a) New York Heart Association functional class ≥ II); *b)* left ventricular ejection fraction > 50% by Simpson method and end-diastolic diameter<60mm; *c)* structural heart disease (left ventricle hypertrophy/left atrial enlargement) and/or diastolic dysfunction estimated by 2-dimensional echocardiography The findings of this trial have been previously published.[11] A total of 47 patients (77% of the total sample) with available plasma samples at enrollment were included in this analysis. The study was registered on ClinicalTrials.gov (Identifier: NCT02638961). Endotrophin was measured in serum.

Randomized trial of vasodilator therapy in HFpEF: We studied participants with HFpEF enrolled in a previous phase IIa trial designed to assess the effect of isosorbide dinitrate, isosorbide dinitrate plus hydralazine or placebo on pulsatile hemodynamics and LV remodeling

in HFpEF[7]. We measured biomarker concentrations using available frozen plasma samples collected during the baseline visit (*n*=45). Inclusion criteria for the trial included symptomatic heart failure with a preserved ejection fraction (LVEF>50%), in addition to at least one of the following: (a) prior hospitalization for decompensated heart failure; (b) acute treatment for heart failure requiring intravenous diuretics or hemofiltration; (c) echocardiographic evidence for elevated filling pressures; d) chronic treatment with a loop diuretic for control of symptoms; (e) or an elevated NT-pro-BNP[12]. Participants needed to be on stable medical therapy for the past month. Exclusion criteria included any rhythm other than sinus with native conduction; non-cardiac conditions that significantly limit exercise (orthopedic or neuromuscular); known hypertrophic, infiltrative, or inflammatory cardiomyopathy; pericardial disease; significant pulmonary disease; primary pulmonary arterial hypertension; acute coronary syndrome or coronary revascularization within the past 60 days; clinically significant perfusion defects on stress imaging without subsequent revascularization; significant valvular disease (e.g. $\geq$ moderate mitral regurgitation or aortic stenosis); uncontrolled hypertension (systolic blood pressure>180 mmHg or diastolic blood pressure >100 mmHg); prior reduced LVEF<50% (i.e., recovered EF); hemoglobin<10g/dL; current therapy with organic nitrates or hydralazine; and elevations in liver function tests. The presence of HFpEF was adjudicated by 2 cardiologists with expertise in HFpEF (JAC and PZ), and individuals with an alternative explanation for symptoms were also excluded. The protocol was approved by the institutional review boards of the Philadelphia Veterans Affairs Hospital and the Hospital of the University of Pennsylvania. All participants provided written informed consent. The study was registered on ClinicalTrials.gov (www.ClinicalTrials.gov; NCT01516346).[13-16]

Aim 3: Cross-sectional comparison of values biomarkers between HFpEF, HFrEF and hypertensive controls

Study 3A: Prospective multicenter biobanking study (n=57): We measured biomarkers using frozen plasma samples from a previously performed prospective biobanking study in which we enrolled 57 participants with HFpEF (n=15), HFrEF (n=27), or hypertension but no HF (n=15), in order to compare various non-invasive phenotypes and establish a biobank for biomarker comparisons between these populations. Detailed inclusion and exclusion criteria for this study are detailed in **Table S1**. Participants with HF were enrolled either as stable outpatients or in the hospital setting, during a HF exacerbation. Participants underwent repeat visits throughout 12 and 24 weeks after enrollment. We analyzed biomarkers of collagen formation and degradation from the 12-week collection (*n*=52; HTN=15; HFpEF=12; HFrEF=25), in order to avoid confounding by the decompensated vs. stable state present upon enrollment. Endotrophin was measured in plasma.

Study 3B: University of Pennsylvania Deep exercise phenotyping study (n=59): This was a cross-sectional analysis of participants with HFpEF, hypertensive individuals without HF symptoms, and healthy controls. The general criteria for participant selection have been previously reported.[17,18] The University of Pennsylvania Institutional Review Board approved the study. All participants provided written informed consent prior to entry.

As reported previously, [17,18] inclusion criteria for HFpEF participants included symptomatic HF (NYHA Class II/III) in the context of a preserved ejection fraction (≥ 50%) and stable medical management for at least 1 month. Participants were required to have evidence of elevated filling pressure which included at least one of the following: (1) Prior admission for HF

requiring intravenous diuretics, (2) history of elevated invasively determined filling pressures (pulmonary capillary wedge pressure >15 mm Hg or left ventricular end-diastolic pressure >16 mmHg), (3) Mitral E/septal e' ratio >15, or (4) Mitral E/e' ratio >8 in addition to one of the following: (a) elevated NT-pro-BNP, (b) left atrial volume index >34 mL/m^2, or (c) chronic use of loop diuretics for control of HF symptoms. Healthy controls were individuals who did not have a history of hypertension or HF. While other cardiovascular conditions were exclusionary, treated hypercholesterolemia was allowed in the healthy group in order to allow representation of elderly participants. Given the near ubiquitous presence of hypertension in HFpEF patients, we enrolled a group of hypertensive individuals without HF symptoms as an additional control group. Hypertensive individuals included those who were treated with antihypertensive medications, had been on stable medical therapy for at least 1 month, and had no history or symptoms consistent with HF. One participant, without a known history of hypertension, was initially enrolled into the healthy group but was found to be hypertensive during the study visit and in the subsequent period afterwards. This participant was therefore included in the hypertensive group, prior to data analysis.

Exclusion criteria were: current atrial fibrillation, inability to exercise, moderate or greater aortic or mitral valve disease, hemoglobin < 10 g/dL, known hypertrophic, inflammatory, or infiltrative cardiomyopathy, pericardial disease, current angina due to clinically-significant obstructive epicardial coronary disease, acute coronary syndrome within the past 2 months, primary pulmonary arterial hypertension, clinically significant lung disease (current use of supplemental oxygen aside from nocturnal O_2 as part of treatment for obstructive sleep apnea; use of steroids or antibiotics within the past 6 months for an acute exacerbation of obstructive pulmonary disease; proximal pulmonary function testing with an FEV_1<50% predicted; 6-minute

walk test with arterial oxygen desaturation), ischemia on stress-testing without subsequent revascularization or demonstration of non-obstructive epicardial coronary disease on coronary angiography, significant liver disease impacting synthetic function or volume control, uncontrolled hypertension (>180/110 mmHg at baseline), eGFR<30 mL/min/m^2 or Cr>2.5 mg/dL, alcohol dependence, or chronic narcotic use that could not be interrupted. Endotrophin was measured in plasma.

Study 3C: Philadelphia VA Medical Center Study (n=320): We prospectively enrolled a convenience sample of adults with HFpEF, HFrEF or no HF at the Corporal Michael J. Crescenz VA Medical Center referred for a cardiac magnetic resonance imaging study. The protocol was approved by the Philadelphia VA Medical Center Institutional Review Board, and all participants provided written informed consent.

In order to optimize case classification according to LVEF and other cardiac parameters detailed below, we measured left ventricular ejection fraction (LVEF) and cardiac structure and function using the current gold-standard method (steady-state free precession cine cardiac MRI). HFrEF was defined as a symptomatic HF in the presence of an LVEF<50%. HFpEF was defined as (1) NYHA Class II-IV symptoms consistent with HF; (2) LVEF>50%; (3) a mitral E wave to annular (e') ratio >14; or at least 2 of the following: (a) a mitral E wave to annular e' ratio >8; (b) treatment with a loop diuretic for control of HF symptoms; (c) left atrial volume index >34 mL/m^2 of body surface area (BSA); (d) NT-pro B-type natriuretic peptide level >200 pg/mL; and (e) LV mass index >149 g/m^2 in men and 122 g/m^2 in women. Participants without HF had an LVEF >50%, and no symptoms and signs consistent with HF. Key exclusion criteria were as follows: (1) Claustrophobia; (2) Presence of metallic objects or implanted medical devices in body; (3) Atrial

fibrillation, flutter or significant arrhythmia at the time of enrollment, which may compromise the study measurements; (4) History of sarcoidosis or amyloidosis, or suspected infiltrative heart disease. Endotrophin was measured in serum.

Section 3: Methods - Statistical analysis – meta-analysis

Individual patient data meta-analysis was performed using a one-stage and two-stage approach. The one-stage meta-analysis used mixed effects survival modeling, which applied a random slop and intercept model to account for cohort-level random effects with a random study intercept and random endothelin effects. A random intercept and slope model with an unstructured covariance structure was used to minimize the correlation between slopes and intercepts.[19] Internal-external cross-validation was performed using a stratified intercept for each study.[20] Model performance was assessed using Harrell's C, the continuous net reclassification index, and integrated discrimination improvement.[21-23]

The two-stage individual patient meta-analysis applied random effects inverse-variance meta-analysis to Cox proportional hazards models with profile likelihood estimation of variance.[24] Heterogeneity was assessed using Cochran's Q test and was quantified by the I^2 index.[25] Missing covariate data were addressed using multiple imputation with chained equations applying iterations of 10 imputed datasets.[26]

Analyses were performed using packages idpmetan, idi, nri, mestreg, and mi impute in STATA version 16.1 (Statacorp LP, College Station, TX).

Acknowledgements

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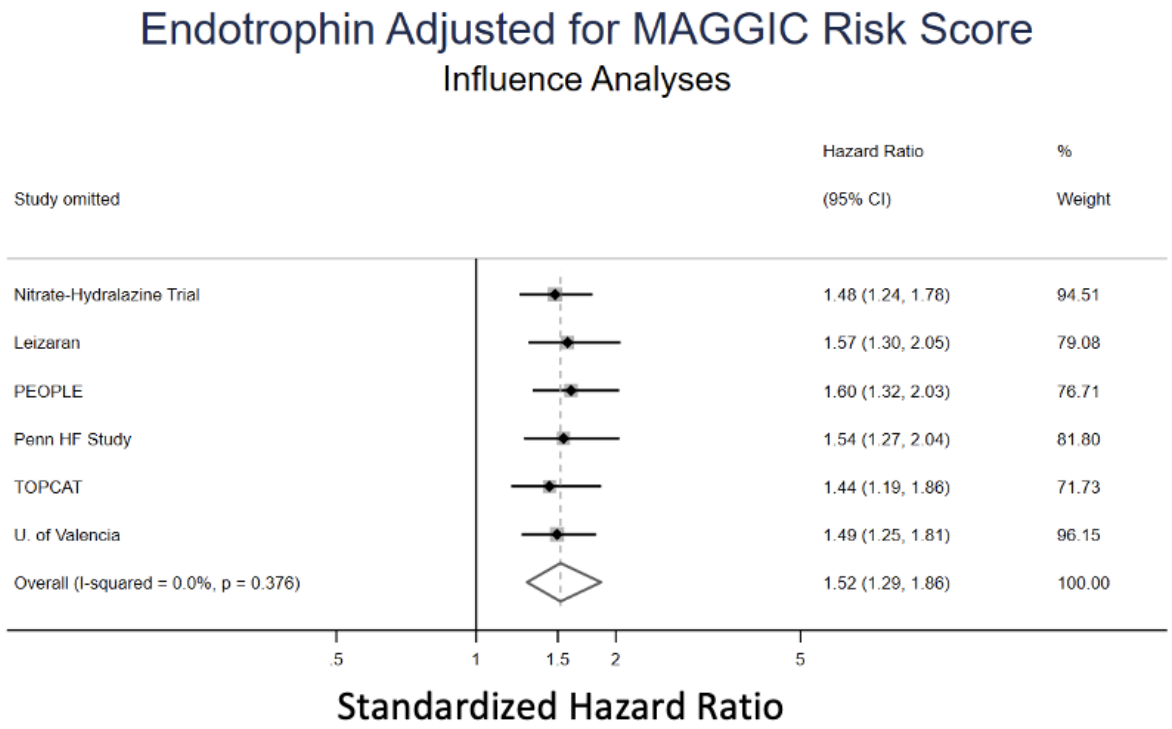
Figure S1. Influence analysis of the various cohorts included in the meta-analysis to assess the relationship between endotrophin and death (A) and death or heart failure hospitalization (DHFA; B) upon removal of each respective cohort from the meta-analysis.

A.



B.

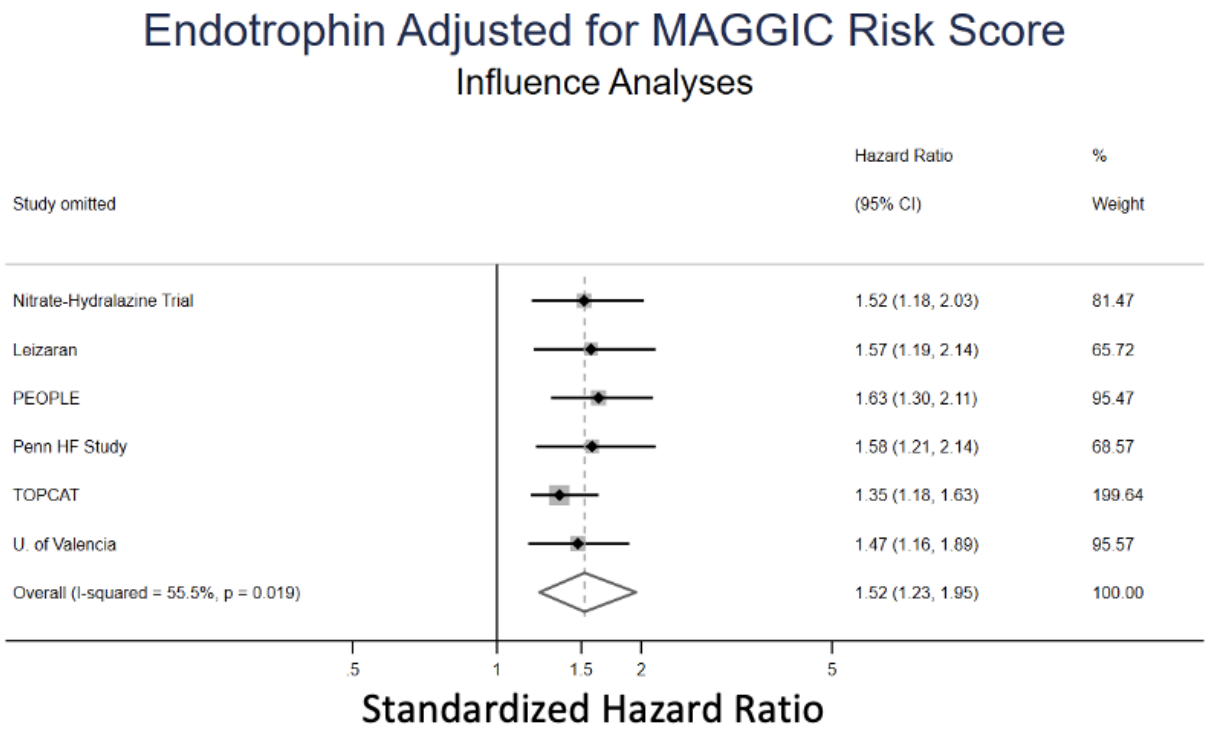
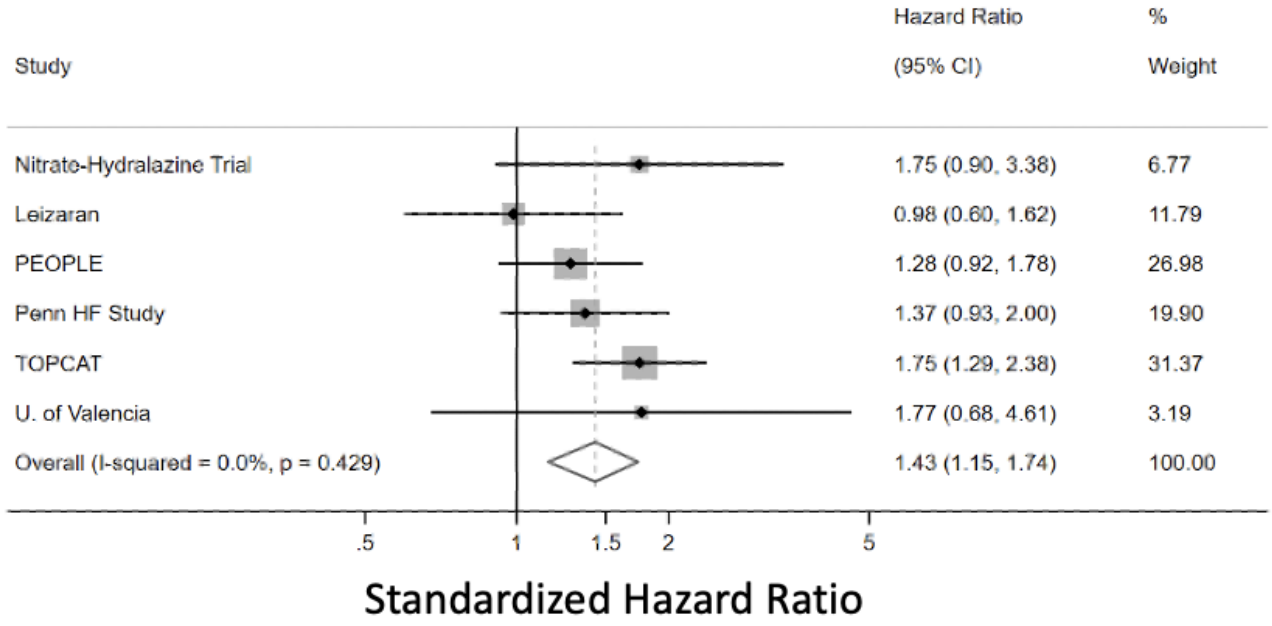


Figure S2. Two step meta-analysis of the relationship between endotrophin and death (A) and death or heart failure hospitalization (B) after adjustment for the MAGGIC risk score and NT-proBNP

A.



B.

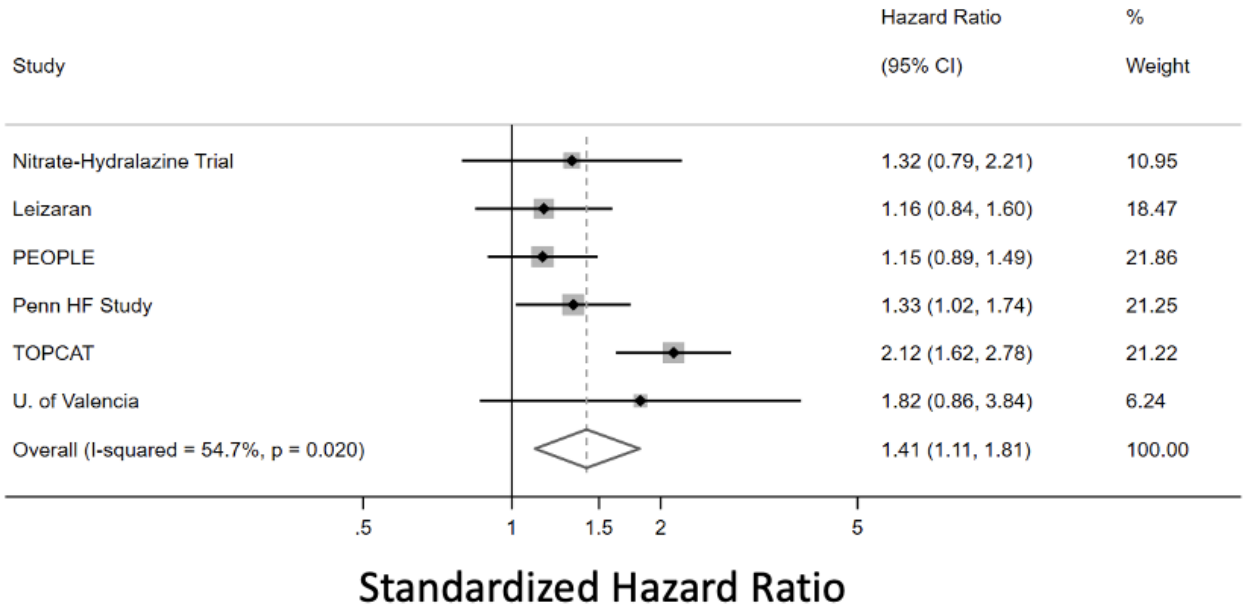
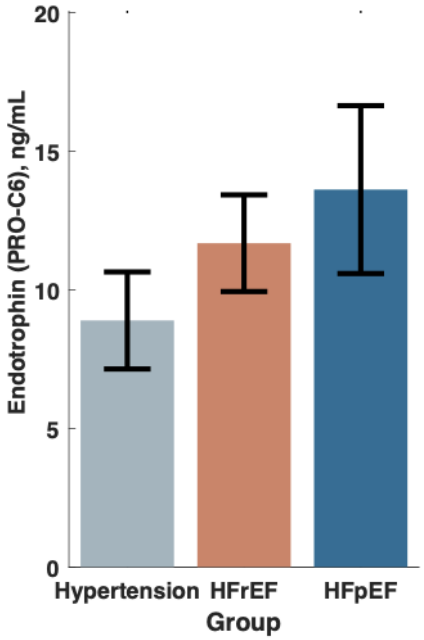
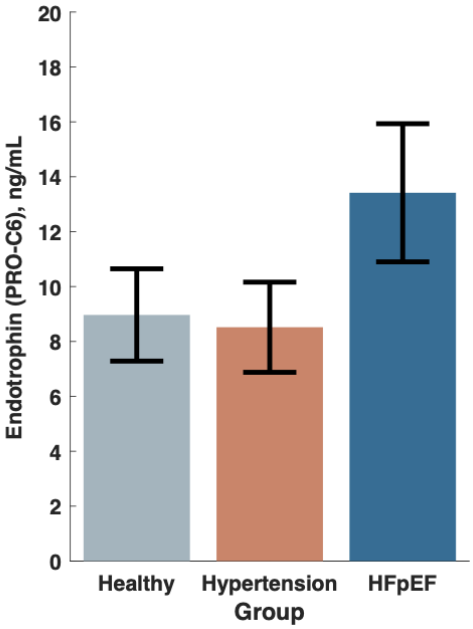


Figure S3. Comparisons of circulating endotrophin between participants with HFpEF, HFrEF and non-HF controls in the multicenter biobanking study (Study 3A, panel A), the University of Pennsylvania deep phenotyping study (Study 3B, Panel B), the Philadelphia VA Medical Center study (Study 3C, Panel C) and HFrEF vs HFpEF participants enrolled in the Penn Heart Failure Study (Panel D). The error bars represent 95% Confidence intervals.

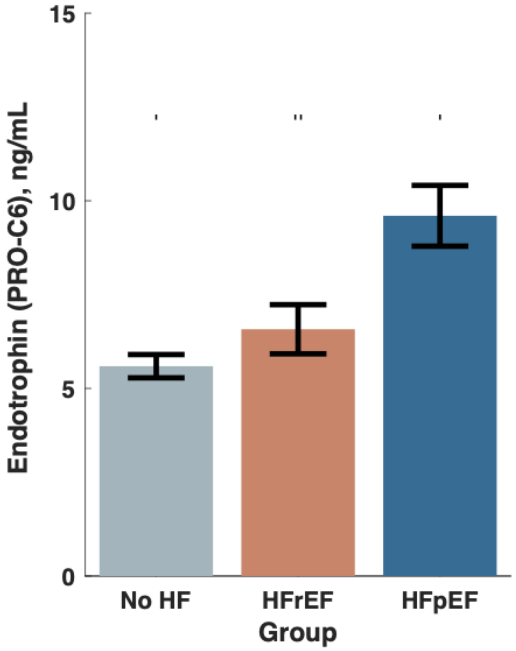
A. Multicenter biobanking study



B. University of Pennsylvania deep phenotyping study



C. Philadelphia VA Medical Center study



D. Penn Heart Failure Study

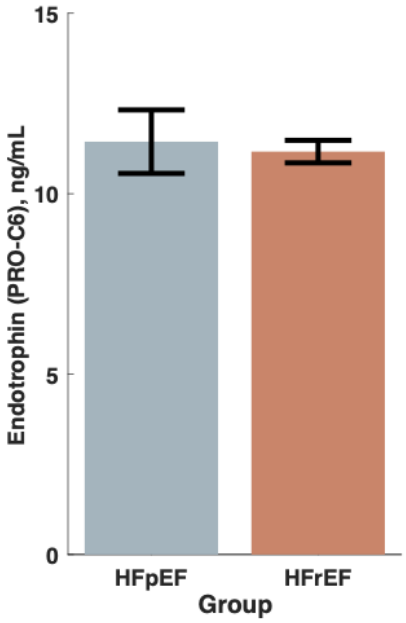


Table S1. Reproducibility and other technical parameters of PRO-C6 kits used for endotrophin measurements in this study. The recovery percentage of the concentration of 5 quality control human serum samples have been evaluated in the different kits. Highest intra- and inter-assay coefficients of variation are also shown, along with the concentration of the standard peptide, two kit controls as well as the slope and IC50 of the standard curve.

Lot#	QC-Panel Recovery (%, 5 samples)	QC-Panel highest Inter Assay CV of single sample (%)	QC-Panel Highest Intra Assay CV of single sample (%)	StdA (ng/ml)	Kit Control 1 (ng/ml)	Kit Control 2 (ng/ml)	Standard curve slope	Standard Curve IC50 (ng/ml)
BL1806A	96.9	16.8	4	102	4.5	14.4	0.91	4.08
BL1808A	98.6	8.2	5.5	115	4.9	14.6	0.92	4.78
BL1810A	96.0	10.8	4.1	122.7	4.8	14.7	0.92	4.53
BL1904A	100	13.7	3.1	127.1	5.2	14.9	0.98	4.4
BL1908A	100.1	14.3	5.3	101.6	5.1	15	1.01	4.94
BL2001A	97.8	7.1	5.4	100.6	3.9	12.4	0.96	4.95
Mean	99.22	11.97	5.24	108.87	4.77	14.61	0.94	4.84
Standard deviation	1.86	3.08	1.91	9.74	0.45	1.27	0.03	1.01

Table S2. Inclusion and Exclusion criteria for our multicenter biobanking study (Aim 3)

Stable HFpEF

1. Diagnosis of heart failure with a documented preserved LVEF ≥ 50% (by echocardiography, cardiac MRI or radionuclide imaging) in at least 1 available study and the absence of any study in the last 2 years showing a LVEF <50% (other than in the setting of acute, rapid atrial fibrillation)
2. Current New York Heart Association Class II-IV symptoms
3. A mitral inflow early diastolic velocity (E) to mitral annular velocity (E/e') ratio ≥15 OR An E/e' ratio > 8 in the presence of either: (1) Left ventricular hypertrophy (defined as LV mass index >95 g/m^2 in women and >115 g/m^2 in men OR when indexed by body height. >60 g/$m^{1.7}$ in women and 80 g/$m^{1.7}$ in men) or (2) Left atrial enlargement (defined as left atrial volume index >34 mL/m2)
4. No recent (within the last 2 months) hospitalization for heart failure

Decompensated HFpEF

1. Patients hospitalized for acute decompensated HF (primary diagnosis) with a documented preserved LVEF >50% (by echocardiography, cardiac MRI or radionuclide imaging) and the absence of any study in the last 2 years showing a LVEF <50% (other than in the setting of acute, rapid atrial fibrillation)
2. Signs of congestion on chest radiograph (at any time during hospitalization)
3. BNP levels >100 ng/L or NT-proBNP levels >300 ng/L (at any time during hospitalization)
4. A mitral inflow early diastolic velocity (E) to mitral annular velocity (E/e') ratio ≥15 OR An E/e' ratio > 8 in the presence of either: (1) Left ventricular hypertrophy (defined as LV mass index >95 g/m2 in women and >115 g/m^2 in men) or (2) Left atrial enlargement (defined as left atrial volume index >34 mL/m^2)
5. Treatment with at least 40 mg i.v. furosemide or its equivalent before Day 1

Stable HFrEF

1. Patients who have a diagnosis of HFrEF with a documented LVEF ≤ 45% (by echocardiography, cardiac MRI or radionuclide imaging by within the past 1 year).
2. Current New York Heart Association Class II-IV symptoms.
3. Stable medical therapy as defined by: (1) No addition or removal of an angiotensin-converting-enzyme (ACE) inhibitor, angiotensin II receptor blocker (ARB), sacubitril/valsartan [EntrestoTM], beta-adrenergic receptor blocker, or calcium channel blockers (CCBs) for 30 days; (2) No change in dosage of ACE, ARBs, beta-blockers or CCBs of more than 100% for 30 days; (3) No change in diuretic dose for at least 10 days.
4. No recent (within the last 2 months) hospitalization for heart failure

Decompensated HFrEF

1. Patients hospitalized for acute decompensated HF (primary diagnosis) with a documented LVEF <45% (by echocardiography, cardiac MRI or radionuclide imaging)
2. Signs of congestion on chest radiograph (at any time during hospitalization)
3. B-type natriuretic peptide (BNP) levels > 100 ng/L or NT-proBNP levels > 300 ng/L (at any time during hospitalization)
4. Treatment with at least 40 mg i.v. furosemide or its equivalent before Day 1

Hypertensive controls

1. Individuals with history of arterial hypertension and on stable (> 4wks) anti- hypertensive therapy
2. Absence of any current or previous symptoms of heart failure (dyspnea on exertion or lower extremity edema, unless these are attributable to be due to calcium channel blocker use or venous insufficiency as per physician's evaluation)
3. No diuretic use for volume overload or lower extremity edema
4. No known elevation of BNP or NT-pro-BNP levels during standard clinical testing

Table S3. Background information on the broader population affected by HF

Condition Under Investigation	Heart Failure
Special considerations related to:	
Sex and gender	Women are more likely than males to have HF with preserved EF (HFpEF) and are substantially less likely to develop HF *with reduced ejection fraction (HFrEF).*
Age	The prevalence of both HFpEF and HFrEF increases steeply with age.
Race or ethnic group	HF affects black persons disproportionately in the US
Geography	Age and cause of HFrEF vary among countries. Patients in Latin America and Asia are younger and more often exhibit non-ischemic etiologies. Less population-based data are available for HFpEF but limited data from randomized trials suggest that patients from Western Europe are older whereas central/Eastern European patients are younger. North American HFpEF patients exhibit the highest prevalence of obesity and diabetes. Latin American patients are younger and exhibit a high prevalence of obesity. Asia-Pacific HFpEF patients exhibit a high prevalence of diabetes despite a relatively low prevalence of obesity.
Other considerations	Additional differences in clinical populations between countries may arise as a result of differences in the awareness and diagnostic workup, particularly for HFpEF
Overall representativeness of this study	Our study includes multiple cohorts which include participants from North America, South America, Europe and New Zealand. In general, the study populations exhibit typical demographic and clinical characteristics of HFpEF and HFrEF, but all of these studies included convenience samples, rather than probabilistic population-based samples.

Table S4. General characteristics of TOPCAT study participants enrolled in the Americas with vs. without available plasma samples. Numbers represent Mean (SD), Median (IQR) or counts (%)

	Participants without available samples (n=1560)	***Participants with available samples (n=205)***	***P value***
Demographic and other Characteristics			
Age, years	72 (64,79)	72 (64,79)	0.9054
Male Sex	770 (49.39%)	113 (54.85%)	0.1405
Race			<0.001
White	1206 (77.36%)	176 (85.44%)	
Black	276 (17.70%)	26 (12.62%)	
Asian	18 (1.15%)	1 (0.49%)	
Other	67 (4.30%)	3 (1.46%)	
BMI, kg/m^2	32.8 (27.9,38.5)	33.1 (28.5,37.8)	0.5387
Heart rate, bpm	68 (61,76)	66.5 (60,76)	0.0456
Systolic BP, mmHg	130 (118,139)	124 (114,136)	0.0039
Diastolic BP, mmHg	70 (62,80)	70 (62,78)	0.0428
Estimated GFR, ml/min/1.73 m^2	60.9 (48.8,76.8)	62.2 (50.5,76)	0.5908
NT-proBNP, pg/mL*	918 (555,2022)	1068 (554,1805)	0.5914
BNP, pg/mL†	261 (149,448)	210 (151,419)	0.3258
Medical History			
NYHA class III-IV	540 (34.70%)	80 (38.83%)	0.2434
Myocardial Infarction	313 (20.09%)	46 (22.33%)	0.4529
Stroke	143 (9.18%)	15 (7.28%)	0.3703
COPD	269 (17.27%)	22 (10.68%)	0.0167
Hypertension	1392 (89.35%)	195 (94.66%)	0.0170
Atrial Fibrillation	640 (41.08%)	102 (49.51%)	0.0212
Diabetes Mellitus	692 (44.42%)	96 (46.60%)	0.5531
Previous Heart Failure Hospitalization	924 (59.27%)	116 (56.59%)	0.4628
Medication Use			
Beta Blockers	1215 (77.98%)	172 (83.50%)	0.0698
Calcium Channel Blockers	600 (38.51%)	81 (39.32%)	0.8225
Diuretics	1385 (88.90%)	187 (90.78%)	0.4153
Glucose-lowering agents	628 (40.31%)	91 (44.17%)	0.2885
ACE Inhibitors or ARBs	1240 (79.59%)	154 (74.76%)	0.1094
Statins	995 (63.86%)	153 (74.27%)	0.0032

* Available in 291 and 67 participants without vs. with available samples for endotrophin measurements, respectively.

† Available in 614 and 84 participants without vs. with available samples for endotrophin measurements, respectively.

Table S5. General characteristics of TOPCAT study participants enrolled in the Americas with available plasma samples stratified by tertiles of endotrophin. Numbers represent Mean (SD), Median (IQR) or counts (%)

	Lowest Tertile	***Mid tertile***	***Highest tertile***
Demographic and other Characteristics			
Age, years	71 (66,78)	72 (63,79)	75 (64,82)
Male Sex	39 (59.09%)	41 (57.75%)	32 (47.06%)
Race			
White	59 (89.39%)	64 (90.14%)	53 (77.94%)
Black	5 (7.58%)	7 (9.86%)	13 (19.12%)
Other	2 (3.03%)	0 (0.00%)	2 (2.94%)
BMI, kg/m^2	31.7 (28.5,36)	33.3 (28.1,38.7)	33.6 (29.1,37.9)
Heart rate, bpm	66.5 (60,72)	66 (60,76)	68 (60,76)
Systolic BP, mmHg	124±14	126±14	124±15
Diastolic BP, mmHg	71.8±10.4	69.4±11.9	67.5±10.1
Estimated GFR, ml/min/1.73 m^2	69.5 (60.8,80.6)	64 (55.4,76.1)	50.3 (38.2,71.1)
Medical History			
NYHA class III-IV	18 (27.27%)	23 (32.39%)	38 (55.88%)
Myocardial Infarction	14 (21.21%)	19 (26.76%)	13 (19.12%)
Stroke	4 (6.06%)	6 (8.45%)	5 (7.35%)
COPD	11 (16.67%)	4 (5.63%)	7 (10.29%)
Hypertension	64 (96.97%)	67 (94.37%)	63 (92.65%)
Atrial Fibrillation	37 (56.06%)	34 (47.89%)	31 (45.59%)
Diabetes Mellitus	19 (28.79%)	38 (53.52%)	38 (55.88%)
Medication Use			
Beta Blockers	57 (86.36%)	59 (83.10%)	55 (80.88%)
Calcium Channel Blockers	24 (36.36%)	28 (39.44%)	29 (42.65%)
Diuretics			
Glucose-lowering agents	18 (27.27%)	36 (50.70%)	36 (52.94%)
Insulin Use	4 (6.06%)	17 (23.94%)	19 (27.94%)
ACE Inhibitors or ARBs	49 (74.24%)	55 (77.46%)	49 (72.06%)
Statins	52 (78.79%)	50 (70.42%)	50 (73.53%)
Endotrophin, ng/mL	9.2 (7.8,10)	13.2 (11.9,14.5)	22.5 (17.9,27.1)

Table S6. Relationship between pro-C6 levels and the incidence of the primary endpoint and of death or HF admission in various models in the TOPCAT trial

Model	Standardized HR for Endotrophin (95%CI)	*P* value	Standardized HR for the MAGGIC Risk Score (95%CI)	*P* value	Standardized HR for BNP (95%CI)	*P* value
Primary Endpoint (NPE=60)						
Unadjusted	2.10 (1.62-2.71)	<0.001	1.22 (0.93-1.60)	0.157	1.45 (1.18-1.78)	<0.001
Adjusted *	2.08 (1.61-2.71)	<0.001	1.05 (0.81-1.37)	0.695	1.15 (0.93-1.42)	0.201
Death (NPE=46)						
Unadjusted	1.74 (1.36-2.24)	<0.001	1.48 (1.07-2.05)	0.018	1.43 (1.13-1.81)	0.002
Adjusted *	1.66 (1.29-2.16)	0.001	1.29 (0.93-1.79)	0.129	1.20 (0.93-1.55)	0.172
Death or HF admission (NPE=72)						
Unadjusted	2.11 (1.67-2.67)	<0.001	1.32 (1.03-1.70)	0.031	1.52 (1.27-1.82)	<0.001
Adjusted *	2.07 (1.63-2.64)	<0.001	1.14 (0.88-1.45)	0.312	1.22 (1.01-1.48)	0.041

*The model included both endotrophin and the MAGGIC risk score
Non-normally distributed parameters were Box-Cox transformed and all parameters are standardized as z scores. The Hazard ratios therefore correspond to a unit increase in the z score of the parameter.
NPE=number of participants who reached the endpoint.

Table S7. Performance of endotrophin compared to existing predictors of HF outcomes: Harrel's C and Somer's D with bootstrapping in the TOPCAT trial

	Harrel's C (95% CI) with endotrophin	Harrel's C (95% CI) without endotrophin	Somers' D (95% CI) with endotrophin	Somers' D (95% CI) without endotrophin
Endotrophin + NT-proBNP + MAGGIC vs. NT-proBNP + MAGGIC alone				
Primary endpoint	0.73 (0.65-0.81)	0.59 (0.56-0.63)	0.43 (0.30-0.55)	0.27 (0.14-0.41)
Death	0.73 (0.65-0.81)	0.68 (0.64-0.72)	0.37 (0.23-0.52)	0.27 (0.13-0.42)
Death or HF hospitalization	0.73 (0.66-0.80)	0.62 (0.59-0.65)	0.43 (0.32-0.54)	0.30 (0.18-0.42)
Endotrophin + MAGGIC vs. MAGGIC alone				
Primary endpoint	0.71 (0.64-0.77)	0.63 (0.61-0.66)	0.41 (0.28-0.54)	0.27 (0.21-0.32)
Death	0.69 (0.61-0.77)	0.66 (0.64-0.69)	0.38 (0.25-0.51)	0.33 (0.28-0.38)
Death or HF hospitalization	0.70 (0.64-0.77)	0.65 (0.63-0.67)	0.41 (0.30-0.52)	0.30 (0.25-0.34)
Endotrophin + NT-proBNP vs. NT-proBNP alone				
Primary endpoint	0.71 (0.64-0.78)	0.60 (0.54-0.66)	0.43 (0.30-0.55)	0.20 (0.08-0.33)
Death	0.67 (0.59-0.75)	0.59 (0.52-0.67)	0.34 (0.18-0.51)	0.19 (0.03-0.34)
Death or HF hospitalization	0.71 (0.65-0.78)	0.61 (0.56-0.66)	0.43 (0.32-0.54)	0.22 (0.11-0.33)
Endotrophin vs. NT-proBNP alone				
Primary endpoint	0.70 (0.64-0.77)	0.60 (0.54-0.66)	0.41 (0.29-0.53)	0.20 (0.08-0.33)
Death	0.67 (0.59-0.76)	0.59 (0.52-0.67)	0.35 (0.19-0.50)	0.19 (0.03-0.34)
Death or HF hospitalization	0.71 (0.64-0.77)	0.61 (0.56-0.66)	0.41 (0.31-0.52)	0.22 (0.11-0.33)

Table S8. General Characteristics of Study Participants included in the various studies in the participant-level meta-analysis stratified by tertiles of endotrophin (Aim 2).
Numbers represent Mean (SD), Median (IQR) or counts (%). For general characteristics of TOPCAT participants included, please refer to Table S5.

Penn Heart Failure Study

	Lowest tertile	Mid-tertile	Highest tertile
Age, years	55.6 (42.2,65)	65.7 (57.7,76.8)	65.3 (56.4,73.7)
Male sex	32 (55.17%)	30 (51.72%)	22 (37.93%)
SBP, mmHg	124 (115,136)	129 (110,142)	126 (117,146)
BMI, kg/m^2	28.6 (25.4,31.6)	34.1 (28.2,39.1)	32 (26.6,43.9)
Smoking	6 (10.34%)	2 (3.45%)	4 (6.90%)
Diabetes	8 (13.79%)	23 (39.66%)	32 (55.17%)
SCr, mg/dL	1 (0.8,1.11)	1.1 (0.96,1.3)	1.64 (1.2,2.07)
LV EF, %	57.8 (52.5,65)	60 (55,65)	63.5 (60,65)
NYHA class III/IV	11 (19.30%)	21 (36.21%)	31 (54.39%)
Beta Blocker	33 (56.90%)	43 (74.14%)	44 (75.86%)
ACEI or ARB	39 (67.24%)	46 (79.31%)	34 (58.62%)
Endotrophin level (ng/mL)	6.5 (5.7,7.4)	11 (9.7,12.2)	21.7 (15.8,27.1)

PEOPLE cohort

	Lowest tertile	Mid-tertile	Highest tertile
Age, years	77.4 (72.3,84.4)	79 (71.2,84.1)	80.5 (75,84.7)
Male sex	23 (42.59%)	33 (56.90%)	27 (48.21%)
SBP, mmHg	132 (114,141)	130 (116,141)	126 (110,139)
BMI, kg/m^2	29.6 (24.4,35.2)	29.6 (25.4,32.6)	27.9 (23.6,32.8)
Smoking	5 (9.26%)	4 (6.90%)	0 (0.00%)
Diabetes	17 (31.48%)	23 (40.35%)	22 (39.29%)
COPD	24 (44.44%)	18 (31.58%)	13 (23.64%)
SCr, mg/dL	0.98 (0.88,1.12)	1.26 (1,1.46)	1.61 (1.24,1.98)
LV EF, %	65.9±7.8	66.4±7.8	65.8±9
NYHA class III/IV	21 (38.89%)	16 (28.07%)	24 (42.86%)
Beta Blocker	40 (74.07%)	40 (68.97%)	42 (75.00%)
ACEI or ARB	43 (79.63%)	41 (70.69%)	41 (73.21%)
Endotrophin level (ng/mL)	9.5 (7.5,10.4)	13.7 (12.7,15.4)	25.8 (21.2,33.6)

Vasodilator Trial

	Lowest tertile	Mid-tertile	Highest tertile
Age, years	62 (59.3,76)	61 (56.8,65.8)	65 (56.3,70.5)
Male sex	11 (73.33%)	11 (73.33%)	12 (80.00%)
SBP, mmHg	133±26	137±13	138±27
BMI, kg/m^2	33.5±6.8	38.8±7.2	38.1±5.1
Smoking	2 (13.33%)	3 (21.43%)	3 (20.00%)
Diabetes	8 (53.33%)	9 (60.00%)	13 (86.67%)
COPD	3 (20.00%)	3 (20.00%)	4 (26.67%)
SCr, mg/dL	0.93 (0.81,1.25)	1.45 (1.18,1.63)	1.54 (1.09,2.48)
LV EF, %	62±5.9	61.1±7.2	64.2±9.5
Beta Blocker	7 (46.67%)	9 (60.00%)	10 (66.67%)
ACEI or ARB	11 (73.33%)	10 (66.67%)	11 (73.33%)
Endotrophin level (ng/mL)	8 (7.4,8.6)	10.8 (9.5,12.6)	19.3 (15.6,27.8)

Leizarán Cohort

	Lowest tertile	Mid-tertile	Highest tertile
Age, years	70 (64,79)	73 (69.3,83)	79.5 (74,84)
Male sex	33 (56.90%)	23 (38.98%)	23 (39.66%)
SBP, mmHg	153 (140,170)	140 (130,159)	145 (130,162)
BMI, kg/m^2	31.2 (27.8,34.7)	30.4 (27.9,34.9)	30.3 (28.4,34.9)
Diabetes	0 (0%)	0 (0%)	0 (0%)
COPD	6 (10.34%)	4 (6.78%)	6 (10.34%)
SCr, mg/dL	0.88 (0.77,0.98)	0.95 (0.83,1.13)	1.16 (0.94,1.5)
LV EF, %	66.3±8.3	64.6±8.7	65.6±8.8
NYHA class III/IV	23 (41.07%)	33 (55.93%)	33 (57.89%)
Beta Blocker	23 (39.66%)	26 (44.07%)	21 (36.21%)
ACEI or ARB	49 (84.48%)	50 (84.75%)	47 (81.03%)
Endotrophin level (ng/mL)	7.1 (5.9,7.8)	9.7 (9.4,10.5)	14.1 (12.6,17.1)

Valencia Cohort

	Lowest tertile	Mid-tertile	Highest tertile
Age, years	72.3±9.3	71.4±8.7	78.1±6.8
Male sex	2 (12.50%)	9 (60.00%)	9 (56.25%)
SBP, mmHg	129±13	132±15	128±14

BMI, kg/m^2	31.9±5.3	32.8±5.5	30.9±5.1
Smoking	1 (6.25%)	1 (6.67%)	1 (6.25%)
Diabetes	4 (25.00%)	8 (53.33%)	9 (56.25%)
COPD	1 (6.25%)	0 (0.00%)	3 (18.75%)
SCr, mg/dL	0.83±0.17	1.16±0.29	1.57±0.51
LV EF, %	67.7±11.3	67.7±9.3	64.3±9.8
NYHA class III/IV	2 (12.50%)	4 (26.67%)	5 (31.25%)
Beta Blocker	13 (81.25%)	13 (86.67%)	15 (93.75%)
ACEI or ARB	4 (25.00%)	3 (20.00%)	2 (12.50%)
Endotrophin level (ng/mL)	7±0.9	10.3±1.3	17.3±3.8

LVEF=LV Ejection fraction. BMI=body mass index; SBP = systolic blood pressure; BMI=body mass index; SCr=serum creatinine; NYHA=New York Heart Association class. COPD=chronic obstructive pulmonary disease; ACEI =Angiotensin Converting Enzyme inhibitor. ARB=angiotensin receptor blocker.

Table S9. Results of one-stage meta-analysis assessing the relationship between endotrophin and death or DHFA

	Hazard Ratio	95% CI
Death		
ProC6	1.92	1.66-2.22
ProC6 + MAGGIC *	1.54	1.32-1.80
ProC6 + MAGGIC + NTProBNP *	1.44	1.22-1.70
Death or Heart Failure Admission		
ProC6	1.69	1.52-1.89
ProC6 + MAGGIC *	1.48	1.31-1.66
ProC6 + MAGGIC + NTProBNP *	1.40	1.24-1.58

* For adjusted models, full covariates were available in 742 participants

Table S10. Results assessing the relationship between endotrophin and death or DHFA, using Cox models stratified by study rather than meta-analysis. The included population is the same as in the meta-analysis

	Hazard Ratio	95% CI
Death		
ProC6	1.93	1.67-2.23
ProC6 + MAGGIC	1.56	1.34-1.83
ProC6 + MAGGIC + NTProBNP	1.37	1.16-1.63
Death or Heart Failure Admission		
ProC6	1.70	1.52-1.90
ProC6 + MAGGIC	1.50	1.33-1.69
ProC6 + MAGGIC + NTProBNP	1.37	1.20-1.56

Table S11. Model Performance for models including the MAGGIC risk (base model) vs. models also including endotrophin for death (top) and death or heart failure hospital admission (DHFA) in the participant-level meta-analysis

Study	C statistic (95% CI) for the base Model (MAGGIC)	C statistic (95% CI) for the base Model + Endotrophin	NRI	IDI
Death				
TOPCAT	0.61 (0.51-0.70)	0.68 (0.61-0.76)	0.44	0.06
PHFS	0.76 (0.63-0.83)	0.78 (0.71-0.84)	0.30	0.02
PEOPLE	0.66 (0.59-0.74)	0.67 (0.59-0.75)	0.22	0.02
Vasodilator HFpEF trial	0.52 (0.31-0.72)	0.79 (0.66-0.91)	0.89	0.15
Leizarán	0.79 (0.72-0.85)	0.79 (0.72-0.86)	0.39	0.02
U. of Valencia	0.72 (0.52-0.93)	0.77 (0.61-0.93)	0.79	0.09
Overall	**0.70 (0.66-0.74)**	**0.74 (0.70-0.77)**	**0.33**	**0.03**
Death and Heart Failure Admission (DHFA)				
TOPCAT	0.57 (0.50-0.65)	0.70 (0.64-0.77)	0.79	0.12w
PHFS	0.62 (0.55-0.69)	0.64 (0.57-0.71)	0.32	0.04
PEOPLE	0.62 (0.55-0.68)	0.62 (0.56-0.68)	0.21	0.01
Vasodilator HFpEF trial	0.56 (0.40-0.72)	0.59 (0.44-0.74)	0.51	0.07
Leizarán	0.73 (0.68-0.78)	0.74 (0.69-0.79)	0.32	0.01
U. of Valencia	0.67 (0.52-0.82)	0.71 (0.61-0.84)	0.71	0.18
Overall	**0.65 (0.61-0.68)**	**0.67 (0.64-0.70)**	**0.27**	**0.03**

Table S12. General characteristics of study participants with HFpEF, HFrEF and hypertensive controls without HF included in the multicenter biobanking study (Study 3A)

	Hypertension	***HFrEF***	***HFpEF***
Age, years	66.6±4.7	64.6±10	64.8±10.7
Male sex	8 (53.33%)	13 (52.00%)	3 (25.00%)
Race			
White	12 (80.00%)	10 (40.00%)	6 (50.00%)
African-American	3 (20.00%)	15 (60.00%)	6 (50.00%)
Heart rate, bpm	64 (57,73)	72 (61.3,79.3)	65 (61.5,70)
Systolic BP, mmHg	131±13	129±19	138±22
Diastolic BP, mmHg	75.7±7.2	74.4±14.2	71.9±8.1
Serum Creatinine, umol/L	75 (64.5,88.8)	91 (80.8,119)	90 (74,123.5)
Serum Potassium, mmol L	4.2 (3.82,4.45)	4.2 (4.07,4.5)	4.3 (4.1,4.6)
Serum Sodium, mmol L	139±2	139±3	139±2
Total Cholesterol, mmol L	4.94±1.11	4.03±1.01	5.01±1.15
HDL Cholesterol, mmol L	1.22 (1.11,1.58)	1.11 (0.96,1.4)	1.4 (1.19,1.89)
LDL Cholesterol, mmol L	3.06±1	2.34±0.86	2.77±1.02
Triglycerides, mmol L	1.14 (0.97,1.65)	0.88 (0.73,1.23)	1.21 (0.96,1.98)
Albumin, g L	44.5±2.9	40±3	41.8±3.7

Numbers represent mean ± SD, median (IQR) or counts (percentage).

Table S13. General characteristics of study participants with HFpEF, hypertensive without HF and normotensive controls enrolled in the University of Pennsylvania phenotyping study (Study 3B)

	Healthy (n=20)	***Hypertensive (n=19)***	***HFpEF (n=20)***
Age, years	54 (39, 63)	66 (50, 71)	67 (62, 76)
Female, n (%)	6 (30)	7 (37)	13 (65)
Race/Ethnicity, n (%)			
White	20 (100)	14 (73.7)	12 (60)
African-American	0 (0)	3 (15.8)	8 (40)
Asian	0 (0)	2 (10.5)	0 (0)
Height, cm	171.9±6.8	171.6±9.7	165.3±9.9
Weight, kg	81.4 (68.7, 85.7)	80.4 (73.0, 89.0)	99.1 (78.3, 113.5)
BMI, kg/m²	26.7 (23.6, 28.7)	27.7 (24.6, 31.5)	32.1 (28.7, 44.4)
Hypertension, n (%)	0 (0)	19 (100)	20 (100)
Diabetes, n (%)	0 (0)	3 (15.8)	11 (55.0)
Insulin	0 (0)	0 (0)	4 (20)
Hyperlipidemia, n (%)	5 (25)	11 (57.9)	18 (90)
OSA, n (%)	1 (5)	4 (21.1)	12 (60)
Beta-Blocker, n (%)	0 (0)	6 (31.6)	16 (80)
CCB, n (%)	0 (0)	7 (36.8)	11 (55)
ACEi/ARB, n (%)	0 (0)	10 (52.6)	14 (70)
Loop diuretic, n (%)	0 (0)	0 (0)	10 (50)
Thiazide diuretic, n (%)	0 (0)	3 (15.8)	4 (20)
Statin, n (%)	3 (15)	8 (42.1)	14 (70)
NYHA Class, n (%)			
II			18 (90)
III			2 (10)
eGFR, mL/min	87.3±12.6	79.5±17.8	71.1±19.9
Hemoglobin, g/dL	13.9±1.3	14.0±1.0	12.7±1.1
NTproBNP, pg/mL	35.0 (17.0, 63.5)	65.0 (34.0, 127.0)	119.0 (49.0, 241.5)
LV Ejection Fraction, %	59.6±6.8	59.8±4.5	61.9±5.6

Numbers represent mean ± SD, median (IQR) or counts (percentage).

ACEi/ARB = angiotensin converting enzyme inhibitor/angiotensin receptor blocker, CCB = calcium channel blocker, OSA = obstructive sleep apnea, SD = standard deviation, TD = tissue Doppler

Table S14. General characteristics of study participants with HFpEF, HFrEF and without HF included in VA study (Aim 3)

	No HF (n=180)	*HFrEF (n=59)*	*HFpEF (n=81)*
Age, years	64 (57,69)	66 (58,71)	64 (58.8,70.3)
Male sex	163 (90.56%)	57 (96.61%)	67 (82.72%)
Race			
White	93 (51.67%)	23 (38.98%)	32 (39.51%)
African American	77 (42.78%)	34 (57.63%)	47 (58.02%)
Other	10 (5.56%)	2 (3.39%)	2 (2.47%)
Body mass index (kg/m^2)	30.2 (25.6,36.1)	28.9 (24.5,32.4)	36.9 (30.1,41.7)
Systolic BP, mmHg	142±18	145±22	146±20
Diastolic BP, mmHg	82.1±11.1	86.7±12.5	84.2±11.6
Current Smoking	47 (26.55%)	13 (22.41%)	15 (18.75%)
Hypertension	130 (72.63%)	46 (77.97%)	73 (90.12%)
Coronary artery disease	45 (25.14%)	25 (42.37%)	28 (34.57%)
COPD	14 (7.87%)	9 (15.25%)	15 (18.52%)
Diabetes mellitus	67 (37.43%)	23 (38.98%)	54 (67.50%)
Estimated GFR	88 (68.3,104)	76 (61.5,95.8)	69 (55,97.3)
NT-proBNP	327 (97,866)	2236 (746,4601)	486 (106,1419)
Medication Use			
Beta blockers	80 (44.69%)	48 (81.36%)	50 (61.73%)
Aspirin	92 (51.40%)	41 (69.49%)	53 (65.43%)
ACEI or ARBs	85 (47.75%)	46 (77.97%)	60 (74.07%)
Statins	111 (62.36%)	41 (69.49%)	54 (66.67%)
Calcium channel blockers	43 (24.16%)	9 (15.25%)	28 (34.57%)
Insulin	25 (13.97%)	7 (11.86%)	31 (38.27%)
Hydralazine	6 (3.35%)	6 (10.17%)	10 (12.50%)
Warfarin	15 (8.43%)	9 (15.25%)	5 (6.17%)
Spironolactone	4 (2.25%)	9 (15.25%)	6 (7.41%)
Long-acting nitrates	14 (7.82%)	10 (17.24%)	20 (24.69%)

Table S15. General characteristics of Penn HF study (PHFS) HFrEF study participants included in the study (n=1642)

	Count (%), Mean, SD, Median (IQR)
Age, years	57.5 (47.2,65.3)
Male sex	1151 (70.10%)
Race	
White	1172 (71.38%)
Black	363 (22.11%)
Asian	17 (1.04%)
Other or unknown	90 (5.48%)
Systolic blood pressure, mmHg	110 (98,124)
Diastolic blood pressure, mmHg	70 (60,78)
Body mass index (kg/m2)	28.5 (25,33.3)
Diabetes mellitus	471 (28.68%)
History of coronary stent	377 (22.96%)
History of coronary artery bypass graft	330 (20.10%)
Atrial fibrillation/flutter	598 (36.42%)
Current smoking	160 (9.74%)
NYHA Class	
I	251 (15.35%)
II	718 (43.91%)
III	545 (33.33%)
IV	121 (7.40%)
Estimated Glomerular filtration rate	57.3 (42.4,71.1)
Left ventricular ejection fraction	25 (17.5,35)
Medication Use	
ACE Inhibitors /Angiotensin receptor blockers	1456 (88.67%)
Aldosterone antagonists	629 (38.31%)
Aspirin	953 (58.04%)
Beta blockers	1494 (90.99%)
Calcium channel blockers	94 (5.72%)
Hydralazine	151 (9.20%)
Nitrate	274 (16.69%)
Statins	853 (51.95%)
Warfarin	673 (40.99%)
Insulin	209 (12.73%)

Table S16. General characteristics of Penn HF Study HFrEF study participants included in the study stratified by tertiles of plasma endotrophin (n=1642).

Numbers represent Count (%), Mean,SD, Median (IQR)

	Lowest Tertile	***Mid-tertile***	***Highest tertile***
Age, years	53.1 (42.7,60.9)	57.6 (47.2,66.1)	61.3 (52.1,68.3)
Male sex	370 (68.27%)	378 (68.48%)	403 (73.54%)
Race			
White	468 (77.23%)	441 (71.94%)	410 (67.21%)
Black	101 (16.67%)	140 (22.84%)	157 (25.74%)
Asian	8 (1.32%)	6 (0.98%)	4 (0.66%)
Other or unknown	29 (4.79%)	26 (4.24%)	39 (6.39%)
Systolic BP, mmHg	112 (100,124)	110 (100,124)	110 (96,124)
Diastolic BP, mmHg	70 (64,78)	70 (60,78)	67 (60,74)
Body mass index (kg/m2)	28 (24.9,31.8)	29.1 (25.1,33.9)	28.9 (25,33.9)
Diabetes mellitus	79 (14.58%)	159 (28.80%)	233 (42.52%)
History of coronary stent	85 (15.68%)	136 (24.64%)	156 (28.47%)
History of coronary artery bypass graft	58 (10.70%)	116 (21.01%)	156 (28.47%)
Atrial fibrillation/flutter	156 (28.78%)	182 (32.97%)	260 (47.45%)
Current smoking	57 (10.52%)	59 (10.69%)	44 (8.03%)
NYHA Class III/IV	140 (25.93%)	217 (39.45%)	309 (56.70%)
Estimated Glomerular filtration rate	67.5 (58,78.4)	58.5 (46.3,69.4)	40.7 (29.4,56.4)
LV ejection fraction	30 (20,42)	25 (20,37.5)	25 (17.5,35)
Medication Use			
ACEI / ARBs	506 (93.36%)	513 (92.93%)	437 (79.74%)
Aldosterone antagonists	187 (34.50%)	236 (42.75%)	206 (37.59%)
Aspirin	280 (51.66%)	327 (59.24%)	346 (63.14%)
Beta blockers	498 (91.88%)	502 (90.94%)	494 (90.15%)
Calcium channel blockers	26 (4.80%)	31 (5.62%)	37 (6.75%)
Hydralazine	16 (2.95%)	41 (7.43%)	94 (17.15%)
Nitrate	41 (7.56%)	74 (13.41%)	159 (29.01%)
Statins	259 (47.79%)	293 (53.08%)	301 (54.93%)
Warfarin	178 (32.84%)	226 (40.94%)	269 (49.09%)
Insulin	29 (5.35%)	61 (11.05%)	119 (21.72%)
Endotrophin, ng/mL	6.6 (5.7,7.4)	10.1 (9.1,11.3)	18.1 (15.1,24.7)

ACEI =Angiotensin Converting Enzyme inhibitor. ARB=angiotensin receptor blocker.BP=blood pressure.

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