

Multiple hormonal and metabolic deficiency syndrome predicts outcome in heart failure: the T.O.S.C.A. Registry

Antonio Cittadini^{1,2,*†}, Andrea Salzano^{3†}, Massimo Iacoviello⁴, Vincenzo Triggiani⁵, Giuseppe Rengo⁶, Francesco Cacciatore¹, Ciro Maiello⁷, Giuseppe Limongelli⁸, Daniele Masarone⁸, Francesco Perticone⁹, Antonio Cimellaro⁹, Pasquale Perrone Filardi^{10,11}, Stefania Paolillo^{10,11}, Antonio Mancini¹², Maurizio Volterrani¹³, Olga Vriz¹⁴, Roberto Castello¹⁵, Andrea Passantino¹⁶, Michela Campo¹⁷, Pietro A. Modesti¹⁸, Alfredo De Giorgi¹⁹, Ines P. Monte²⁰, Alfonso Puzzo²¹, Andrea Ballotta²², Roberta D'Assante^{1,2}, Michele Arcopinto^{1,2}, Paola Gargiulo¹⁰, Angela Sciacqua⁹, Dario Bruzzese²³, Annamaria Colao²⁴, Raffaele Napoli¹, Toru Suzuki²⁵, Kim A. Eagle²⁶, Hector O. Ventura²⁷, Alberto M. Marra^{1,2,28‡}, and Eduardo Bossone^{10,29‡}; On behalf of the T.O.S.C.A. Investigators[¶]

¹Department of Translational Medical Sciences, Federico II University, Via Sergio Pansini, 5, 80131 Naples, Italy; ²Italian Clinical Outcome Research and Reporting Program (I-CORR), Naples, Italy; ³IRCCS SDN, Diagnostic and Nuclear Research Institute, Naples, Italy; ⁴Cardiology Unit, Department of Medical and Surgical Sciences, University of Foggia, 71122 Foggia, Italy; ⁵Interdisciplinary Department of Medicine, Section of Internal Medicine, Geriatrics, Endocrinology and Rare Diseases, University of Bari 'A Moro', Bari, Italy; ⁶Istituto Clinico Scientifici Maugeri SpA Società Benefit (ICS Maugeri SpA SB), IRCCS, Scientific Institute of Telesse Terme, Telesse BN, Italy; ⁷Heart Transplantation Unit, Monaldi Hospital, Azienda Ospedaliera dei Colli, Naples, Italy; ⁸Division of Cardiology, Monaldi Hospital, Azienda Ospedaliera dei Colli, University of Campania Luigi Vanvitelli, Caserta, Italy; ⁹Department of Medical and Surgical Sciences, University Magna Graecia of Catanzaro, Catanzaro, Italy; ¹⁰Department of Advanced Biomedical Sciences, Federico II University, Naples, Italy; ¹¹Mediterranea Cardiocentro, Naples, Italy; ¹²Operative Unit of Endocrinology, Catholic University of the Sacred Heart, Rome, Italy; ¹³Department of Medical Sciences, IRCCS San Raffaele Pisana, Rome, Italy; ¹⁴Heart Center Department, King Faisal Hospital & Research Center, Riyadh, Kingdom of Saudi Arabia; ¹⁵Division of General Medicine, Azienda Ospedaliera Universitaria Integrata, Verona, Italy; ¹⁶Scientific Clinical Institutes Maugeri, IRCCS, Pavia, Italy; ¹⁷Department of Medical and Surgical Sciences, Unit of Endocrinology and Metabolic Diseases, University of Foggia, Foggia, Italy; ¹⁸Dipartimento di Medicina Sperimentale e Clinica, Università degli Studi di Firenze, Florence, Italy; ¹⁹Department of Medical Sciences, School of Medicine, Pharmacy and Prevention, University of Ferrara, Ferrara, Italy; ²⁰Department of General Surgery and Medical-Surgery Specialties, University of Catania, Catania, Italy; ²¹IRCCS Oasi Maria SS, Troina, Enna, Italy; ²²IRCCS Policlinico San Donato, San Donato Milanese, Milan, Italy; ²³Department of Public Health, Federico II University, Naples, Italy; ²⁴Clinical Medicine and Surgery Department, Federico II University, Naples, Italy; ²⁵Department of Cardiovascular Sciences and NIHR Leicester Biomedical Research Centre, University of Leicester, Glenfield Hospital, Leicester, UK; ²⁶Michigan Frankel Cardiovascular Center, University of Michigan, Ann Arbor, MI, USA; ²⁷John Ochsner Heart and Vascular Institute, Ochsner Clinical School, The University of Queensland School of Medicine, New Orleans, LA, USA; ²⁸Center for Pulmonary Hypertension, Thoraxclinic at Heidelberg University Hospital, Heidelberg, Germany; and ²⁹Cardiology Division, A Cardarelli Hospital, Naples, Italy

Received 16 October 2020; revised 4 January 2021; editorial decision 26 January 2021; accepted 2 February 2021; online publish-ahead-of-print 7 March 2021

Aims

Recent evidence supports the occurrence of multiple hormonal and metabolic deficiency syndrome (MHDS) in chronic heart failure (CHF). However, no large observational study has unequivocally demonstrated its impact on CHF progression and outcome. The T.O.S.C.A. (Trattamento Ormonale nello Scompenso CArdiaco; Hormone Treatment in Heart Failure) Registry has been specifically designed to test the hypothesis that MHDS affects morbidity and mortality in CHF patients.

* Corresponding author. Tel/Fax: +39 081 7464375, Email: antonio.cittadini@unina.it

† The first two authors contributed equally to the study.

‡ The last two authors contributed equally to the study.

¶ A complete list of the T.O.S.C.A. Investigators is provided at the end of the paper.

© The Author(s) 2021. Published by Oxford University Press on behalf of the European Society of Cardiology.

This is an Open Access article distributed under the terms of the Creative Commons Attribution Non-Commercial License (<http://creativecommons.org/licenses/by-nc/4.0/>), which permits non-commercial re-use, distribution, and reproduction in any medium, provided the original work is properly cited. For commercial re-use, please contact journals.permissions@oup.com

Methods and Results

The T.O.S.C.A. Registry is a prospective, multicentre, observational study involving 19 Italian centres. Thyroid hormones, insulin-like growth factor-1, total testosterone, dehydropianoandrosterone sulfate, insulin resistance, and the presence of diabetes were evaluated. A MHDS was defined as the presence of ≥ 2 hormone deficiencies (HDs). Primary endpoint was a composite of all-cause mortality and cardiovascular hospitalizations. Four hundred and eighty heart failure patients with ejection fraction $\leq 45\%$ were enrolled. MHDS or diabetes was diagnosed in 372 patients (77.5%). A total of 271 events (97 deaths and 174 cardiovascular hospitalizations) were recorded, 41% in NO-MHDS and 62% in MHDS ($P < 0.001$). Median follow-up was of 36 months. MHDS was independently associated with the occurrence of the primary endpoint [hazard ratio 95% (confidence interval), 1.93 (1.37–2.73), $P < 0.001$] and identified a group of patients with a higher mortality [2.2 (1.28–3.83), $P = 0.01$], with a graded relation between HDs and cumulative events ($P < 0.01$).

Conclusion

MHDS is common in CHF and independently associated with increased all-cause mortality and cardiovascular hospitalization, representing a promising therapeutic target.

Trial registration

ClinicalTrials.gov identifier: NCT023358017

Keywords

Heart failure • Anabolic deficiency • Multiple hormonal and metabolic deficiency syndrome • Hormones • Prognosis • TOSCA

Introduction

Chronic heart failure (CHF) is a major healthcare issue with increasing prevalence, huge estimated cost, and poor prognosis, still approaching a 5-year mortality of 50%.^{1,2} Given such disheartening statistics, an imperative need is to search for novel therapeutic approaches capable of slowing disease progression and improving survival.^{3,4} In this regard, the neurohormonal model, conjecturing CHF pathophysiology as sustained by an excessive activation of numerous pathways, including the sympathetic, renin–angiotensin–aldosterone, and cytokine systems, provided a theoretical framework supporting the actual therapy.^{3,4} Such an approach, although partially successful, has not fulfilled all the promises, and CHF prognosis remains modest.^{1,2} However, to complement the paradigm of neurohormonal activation, a concomitant reduction of anabolic hormonal axes seems to potentially play an important role in CHF progression and prognosis.⁵ Specifically, the so-called multiple hormonal and metabolic deficiency syndrome (MHDS) encompasses several anabolic systems that are down-regulated or impaired in CHF: the somatotrophic axis [including growth hormone (GH) and its tissue effector insulin-like growth factor-1 (IGF-1)], anabolic steroids (testosterone and DHEA-S), and thyroid hormones.^{5–9} Notably, MHDS does not appear to be a mere marker of disease progression, insofar as each defect is associated with impaired clinical status, functional capacity, and increased mortality.^{5–10} In addition, insulin resistance (IR), with or without coexisting overt hyperglycaemia and type 2 diabetes (T2D), appears to have an independent role in the pathogenesis and prognosis of CHF.¹¹ Indeed, in previous preliminary report investigating the impact of hormone deficiencies (HDs) in CHF, the role of IR has been ignored, and the effect of T2D disregarded. Furthermore, promising results observed in recent trials investigating the effect of antidiabetic drugs (i.e. gliflozins) support the idea that hormonal and metabolic players might be involved in the progression of CHF.¹² Despite these premises, to date no large study has focused on the relative role played by MHDS in the

progression and survival of patients with CHF. In particular, no registry-based study has been specifically designed to evaluate the overall impact of HDs on CHF morbidity and mortality.

Aims of the T.O.S.C.A. (Trattamento Ormonale nello Scompenso CARDiaco; Hormone Treatment in Heart Failure, NCT02335801) Registry were to investigate in a prospective multicentre observational registry the prevalence, the clinical significance, and the prognostic impact of HD, alone or combined (i.e. MHDS) in CHF.¹³

Methods

Study population

Study design and population baseline characteristics have been previously described.¹³ In brief, the T.O.S.C.A. Registry is a prospective multicentre observational study recruiting consecutive stable CHF patients with left ventricular ejection fraction (LVEF) $\leq 45\%$, without history of recent acute decompensation, acute coronary syndrome (< 6 months), severe liver (Cirrhosis Child-Turcotte-Pugh B-C), and/or kidney disease (creatinine level > 2.5 mg/dL) or active malignancy, on stable medications for at least 3 months, including any beta-blocker (started at least 6 months before entering the study). Patients with history of current hormonal treatment or overt endocrine diseases were excluded.

Study outcomes

The primary endpoint was a composite of all-cause mortality or cardiovascular hospitalization. Information regarding clinical outcome was obtained directly from patients or their relatives and an independent endpoint committee adjudicated the outcome. Secondary endpoint was the delta change in maximal oxygen consumption (peak VO_2) from baseline, when a follow-up cardiopulmonary exercise test was performed (not mandatory, based on clinical judgement and centre availability).

Study procedures and statistical plan

Study procedures, previously described,¹³ are available as [Supplementary material online](#). In brief, blood samples were collected by venepuncture

after overnight fast. To obtain serum and plasma, samples were centrifuged within 30 min, frozen, and stored at -80°C until assayed. Brain natriuretic peptide levels were assessed using a point-of-care device (RapidPIATM, Sekisui Medical Co, Tokyo, Japan) in a dedicated core-lab (John and Lucille van Geest Biomarker Facility, University of Leicester, UK). Single HD was defined as described in Table 1. Patients were divided in two groups: (i) patients with one or no HD (NO-MHDS) and (ii) patients with two or more HDs (MHDS). All analyses were performed in the entire population and in the subgroup of patients without T2D. The effect of T2D *per se* on the primary endpoint was also evaluated.

Statistical analysis

Normally distributed continuous variables were expressed as mean \pm standard deviation, whereas continuous data with skewed distributions were expressed as median (interquartile range). Categorical variables were expressed as counts and percentages. The distribution of the variables was tested with Kolmogorov–Smirnov test.

The association between analysed variables and survival were established by using Cox proportional hazard regression analyses. Both univariate and multivariable linear models were used to assess potential predictors of survival.

Cox proportional hazards model was also carried out to estimate the Akaike Information Criterion (AIC) for each HD and set up a model useful to find relevant interaction between HD and covariates.

With regard to anabolic status, we considered single HD, the sum of HD detected (from 0 = no deficiencies to ≥ 3 deficiencies) or the presence of an MHDS.

For the multivariable analyses, established predictors of poor outcome in heart failure were employed as variables (i.e. age, sex, aetiology, New York Heart Association (NYHA) class, LVEF, obesity, impaired haemoglobin levels, atrial fibrillation, impaired estimated glomerular filtration rate (eGFR), and B-type natriuretic peptide (BNP); peak VO_2 has been excluded because not available for all patients). Two different models

were used to test the primary endpoint: (i) Model 1: diagnosis of MHDS; (ii) Model 2: patients stratified for number of HD. Results were as hazard ratio (HR) with 95% confidence intervals for a unit increase in the predictor value.

Kaplan–Meier curves for cumulative survival were constructed to assess the impact of severity of anabolic deficiency on endpoints. Differences in event rates between the groups were compared with the Cox-Mantel log-rank test. Schoenfeld residual test was performed to test the proportional hazards assumption for each covariate included in the Cox regression model.

Results

Study population

From the original cohort of 526 patients enrolled in 19 participating centres from April 2013 to July 2017,¹⁴ complete hormonal data were available for 480 patients, who were used for the present analysis. No patient was lost to follow-up. Demographic characteristics at baseline of the final cohort are described in Table 2. No differences were found with regard to heart failure treatment and follow-up duration between the two groups. There were no significant differences (including heart failure treatment and duration of follow-up) between patients included and not included in the present analysis (data not shown). Overall, MHDS (considering T2D or Homeostatic model assessment of insulin resistance—HOMA-IR as one combined deficiency) was diagnosed in 358 patients (74.6%). Specifically, 5.8% displayed no HD, 19.6% only one HD, 32.9% two HD, 27.9% three HD, and 13.8% four or more HD. When patients with T2D were excluded from the analysis, among the remaining 360 patients, 8% displayed no HD, 22% only one HD, 34% two HD, 27% three HD, and 10% four or more HD.

Table 1 Definition of hormone deficiencies and their prevalence

Hormone deficiency	Value for diagnosis	prevalence of hormone deficiencies		
		Total population (N = 480)	Men (N = 386)	Women (N = 94)
IGF-1 deficiency	Serum IGF-1 levels below the 33rd percentile of an age-matched healthy control population: <ul style="list-style-type: none"> • age < 55 years: 122 ng/mL • 55 years < age < 64.9 years) 109 ng/mL; • 65 years < age < 74.9 years: 102 ng/dL • age > 75 years: 99 ng/dL 	221 (46%)	171 (44.3%)	50 (53.2%)
Testosterone deficiency	Serum testosterone levels lower than 300 ng/dL in male or 25 ng/dL in female	204 (42.5%)	174 (45.1%)	30 (31.9%)
DHEAS deficiency	Serum DHEA-S levels lower than 80 $\mu\text{g/dL}$	314 (65.4%)	241 (62.4%)	73 (77.7%)
Insulin-resistance	Type 2 diabetes mellitus (T2D) or Homeostasis Model Assessment (HOMA) greater than 2.5 (HOMA= insulin (mcU/mL) \times glucose (mmol/L)/22.5	308 (64.2%)	244 (63.2%)	64 (68.1%)
Low T3 syndrome	Serum free T3 lower than 3.1 mmol/L with TSH levels within normal range	33 (6.9%)	24 (6.2%)	9 (9.6%)

Serum hormones were analysed in a centralized core-lab (IRCCS SDN, Naples, Italy). Insulin and insulin growth factor-1 (IGF-1) were assayed by an enzyme-labelled chemiluminescent immunometric assay (IMMULITE 2000; IGF-1, interassay CV= 5.7%, Siemens Medical Solutions Diagnostics). Total testosterone was measured with a DPC Coat-A-Count RIA kit. DHEA-S was measured by a solid-phase, competitive chemiluminescent enzyme immunoassay.

Table 2 Demographic characteristics at baseline of final cohort

Variables	Cohort (n = 480)
Age (years)	63.7 ± 11.5
Sex (% male)	80.4
NYHA (% I/II/III/IV)	11/54/33/2
Aetiology (% ischaemic)	52.7
Yr. of disease	7 (2–12)
Systolic blood pressure (mmHg)	121 ± 17
Diastolic blood pressure (mmHg)	74 ± 10
Type 2 diabetes mellitus, n (%)	120 (25)
BMI (kg/m ²)	28.6 ± 5.4
eGFR (mL/min per 1.73 m ²)	86 ± 41
NT pro BNP (pg/mL)	909 (284–2521)
Left ventricular EF (%)	32.3 ± 7.2
Atrial fibrillation (%)	11.2
ICD (%)	36
CRT (%)	11.7
Medication (%)	
B-blocker	87.5
ACE-I/ARBs	86
MRA	39
Diuretics	69.4
Amiodarone	15.8
Digoxin	9.2
Antiplatelets	49
Antithrombotic	27.9
Lipid-lowering medications	51.9
Ivabradine	11
Antidiabetics	15.2
Insulin	10.6

ACE-I, angiotensin-converting-enzyme inhibitors; ARBs, angiotensin-receptor blockers; BMI, body mass index; CRT, cardiac resynchronization therapy; EF, ejection fraction; eGFR, estimated glomerular filtration rate (CKD-EPI); ICD, implantable cardioverter-defibrillator; NYHA, New York Heart Association; NT pro BNP, N-terminal proB-type natriuretic peptide.

Primary endpoint

At the end of the follow-up (patient-average follow-up of 36 months), in the entire population, 271 patients (56%) experienced the primary endpoint (97 deaths—20% and 174 cardiovascular hospitalizations—36%). As shown in *Figure 1A*, 62% of patients in the MHDS group (221 events: 82 deaths—23% and 139 cardiovascular hospitalizations—39%), compared with only 41% of patients in the NO-MHDS group (50 events: 15 deaths—12% and 35 cardiovascular hospitalizations—29%), reached the primary endpoint, resulting in a 50% increase of patients with an event in the MHDS group ($P < 0.001$). With regard to T2D patients, primary endpoint was reached in 70% of the patients with diabetes and 52% in the patients without diabetes ($P = 0.001$). When the 120 patients with diabetes were excluded from the analysis, 34% of patients in the NO-MHDS group ($n = 107$) (36 events: 11 deaths—10% and 25 cardiovascular hospitalizations—23%) and 60% of patients in the MHDS group ($n = 253$) (151 events: 52 deaths—52% and 99 cardiovascular hospitalizations—39%)

experienced the primary endpoint ($P < 0.001$), with an increased risk of outcome of more than 75% in the MHDS. When we performed the analysis in T2D patients ($n = 120$), 90% of the 31 MHDS patients experienced the primary endpoint, as compared with only 75% in the 89 NO-MHDS ($P < 0.01$).

Prognosis

Single predictors

In the univariate Cox proportional hazard regression analyses (*Figure 2*), the following variables were associated with mortality and cardiovascular hospitalization: age, advanced NYHA classes, LVEF, BNP, and anaemia. With regard to specific HD, testosterone deficiency (TD) [1.59 (1.25–2.01), $P < 0.001$], DHEAS deficiency (DHEAS-D) [1.40 (1.07–1.81), $P = 0.011$], IGF-1 deficit [1.43 (1.13–1.82), $P = 0.003$], and IR impairment [i.e. patients with abnormal HOMA index or T2D [1.34 (1.03–1.73), $P = 0.03$] were associated with the primary outcome. Furthermore, when the analysis was performed separately for the presence of HOMA-IR or diabetes, only T2D was associated with the primary endpoint [1.55 (1.20–2.01), $P < 0.001$], whereas HOMA-IR alone, did not show an increased risk of mortality or cardiovascular hospitalization [0.94 (0.74–1.2), $P = 0.064$]. The number of HD [1.35 (1.21–1.52), $P < 0.001$] as well as the presence of MHDS [2.09 (1.52–2.88), $P < 0.001$] were associated with primary outcome. In particular, MHDS identified a group of patients with both higher mortality [2.2 (1.28–3.83), $P = 0.01$] and cardiovascular hospitalization [1.81 (1.29–2.54), $P = 0.001$] (*Figure 1B and C*). When the analysis was performed without the diabetic patients, MHDS was still associated with an increased risk of primary endpoint [HR 2.15 (1.49–3.09), $P < 0.01$], as well as when we performed the analysis only in T2D patients ($n = 120$) [HR 1.87 (1.15–3.05), $P < 0.01$].

Kaplan–Meier analysis performed across HD numbers showed that patients without HD had the best survival rate when compared with those with 1, 2, and ≥ 3 HD, with a graded relation between event rate and number of HD (*Figure 3*). Similar results were obtained when diabetic patients were excluded from the analysis: the presence of increasing number of HD was associated with an increased risk of primary outcome [one HD: HR 1.43 (0.65–3.13), $P = 0.37$; two HD: HR 2.53 (1.22–5.27), $P < 0.01$; HD ≥ 3 : HR 3.08 (1.49–6.38), $P < 0.002$].

Multivariate analysis

MHDS

In the multivariable Cox proportional hazard regression analyses for Model 1-full model (*Figure 4A*), the presence of MHDS was significantly associated with the primary endpoint when adjusted for age, sex, NYHA class, aetiology, LVEF, BNP, and the presence of obesity, impaired eGFR, atrial fibrillation, and anaemia [1.74 (1.21–2.5), $P = 0.003$]. In this model, also sex [1.47 (1.01–2.15), $P = 0.045$], age [1.02 (1.00–1.03), $P = 0.023$], and BNP [1.17 (1.04–1.31), $P = 0.007$] were significantly associated with the outcome. In the multivariable Cox proportional hazard regression analyses for Model 2-simplified (*Figure 4B*), the presence of MHDS was significantly associated with the primary endpoint when adjusted for age, sex, NYHA class, aetiology, LVEF, and BNP [1.93 (1.37–2.73), $P < 0.001$]. When T2D patients were excluded, MHDS remained strongly and significantly associated with the primary endpoint [1.95 (1.31–2.90), $P < 0.001$].

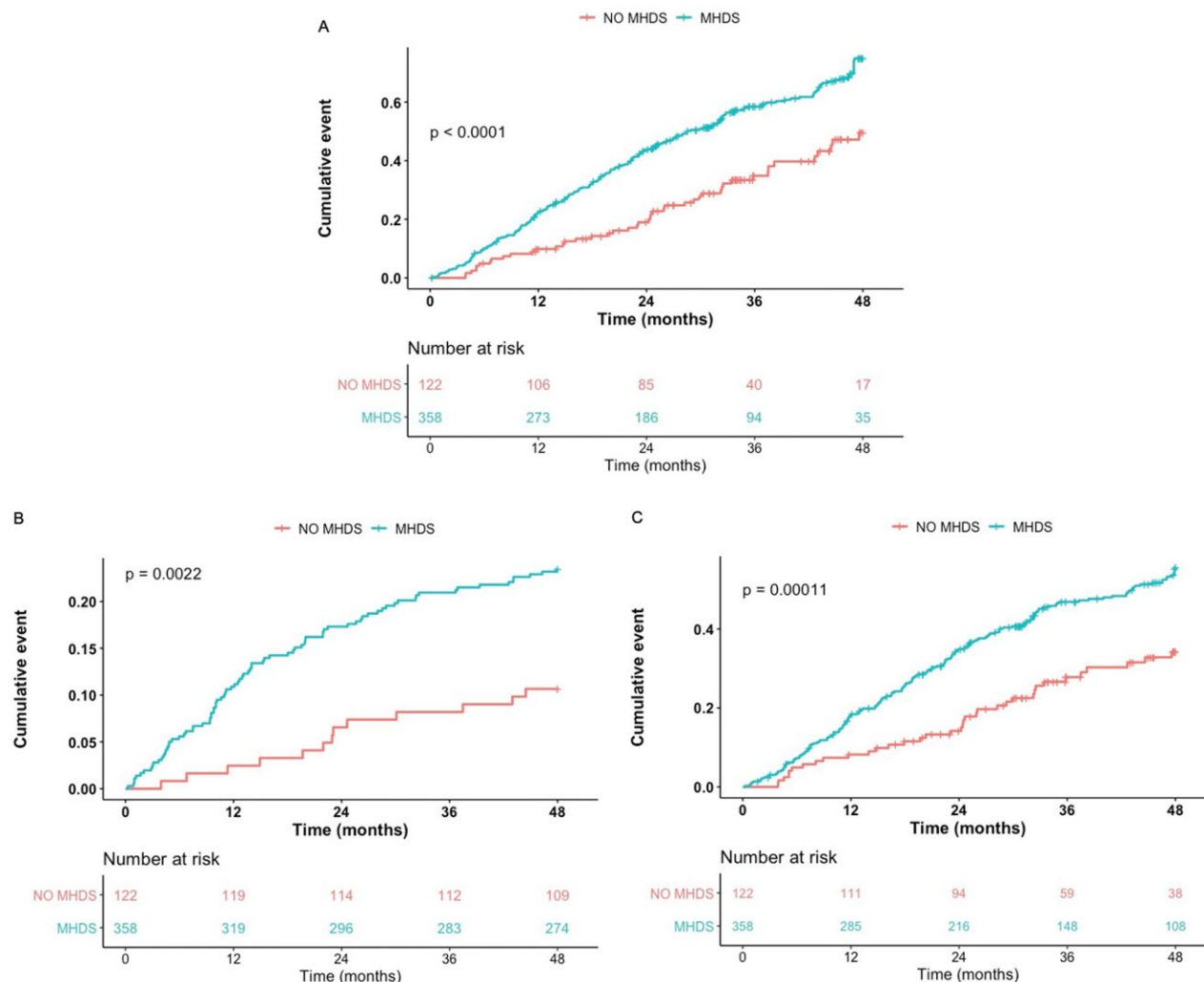


Figure 1 Occurrence of the primary endpoint in patients with MHDS. Kaplan–Meier analysis of composite of all-cause mortality and cardiovascular hospitalizations [1.93 (1.37–2.73), $P < 0.001$] (primary endpoint, A), all-cause mortality [1.02 (1.01–1.03), $P = 0.001$] (B), and cardiovascular hospitalization [1.02 (1.01–1.03), $P = 0.001$] (C), in patients with one or no hormonal deficiencies (NO-MHDS, $n = 122$) vs. patients with two or more hormonal deficiencies (MHDS, $n = 358$). Log-rank testing was applied for calculation of P -values. MHDS, multiple hormonal and metabolic deficiencies syndrome.

The presence of T2D was also significantly associated with the primary endpoint when adjusted for age, sex, NYHA class, aetiology, LVEF, and BNP [1.4 (1.05–1.85), $P = 0.02$].

Schoenfeld residual test confirmed the proportional hazards assumption for each covariate included in the Cox regression model ($\chi^2 = 17.54$, $P = 0.1$).

Number of HD

With regard to Model 3 (Figure 4C), the number of HD detected was significantly associated with outcome even when adjusted for age, sex, NYHA class, aetiology, LVEF, and BNP [1.28 (1.13–1.43), $P < 0.001$]. When diabetic patients were excluded, the presence of increasing number of HD remained associated with an increased risk of primary outcome [one HD: HR 1.72 (0.71–4.22), $P = 0.23$; two

HD: HR 2.85 (1.23–6.62), $P < 0.01$; HD ≥ 3 : HR 3.1 (1.33–7.23), $P < 0.01$].

Prognosis related to specific HD

Association between the presence of a specific single HD at baseline and outcomes was investigated. Kaplan–Meier analyses were performed across TD, DHEA-S D, low IGF-1, HOMA-IR, T2D, and low T3 for composite endpoint from enrolment (Supplementary material online, Figures S1 and S2).

The presence of TD, DHEA-S D, low IGF-1, or T2D, were independently associated with outcome ($P < 0.001$, $P = 0.005$, $P < 0.001$, and $P < 0.001$, respectively). Patients without HD had the best survival rate when compared with those with HD. Low T3 syndrome was not significantly associated with the primary endpoint. However, despite the small number of patients with this hormone defect and

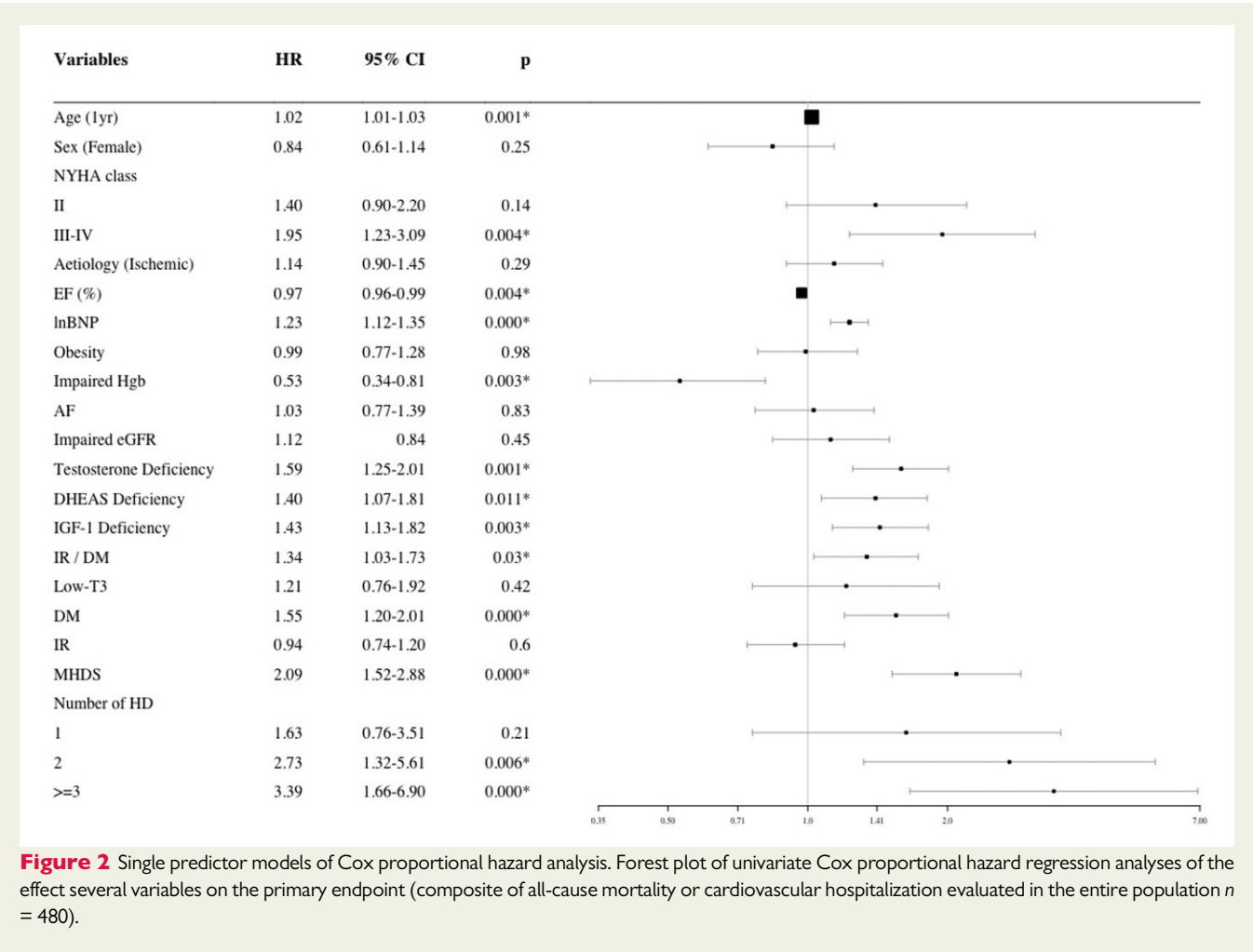


Figure 2 Single predictor models of Cox proportional hazard analysis. Forest plot of univariate Cox proportional hazard regression analyses of the effect several variables on the primary endpoint (composite of all-cause mortality or cardiovascular hospitalization evaluated in the entire population *n* = 480).

the consequent relatively low event rate, in a sub-analysis performed up to 30 months, there was a significant association with the composite of all-cause mortality and cardiovascular hospitalization (*P* = 0.02). No interactions were found between the five HD considered, suggesting a similar role of each HD in determining the primary endpoint.

Cardiopulmonary performance and echocardiographic findings

At baseline, patients with MHDS displayed an impaired cardiopulmonary performance compared with NO-MHDS patients, as shown by significantly lower peak VO₂ (15.5 ± 3.8 and 17.7 ± 4.5 mL/kg/min, respectively, *P* < 0.01) and VO₂ at threshold (10.2 ± 2.8 and 12.1 ± 3.2 mL/kg/min, respectively, *P* < 0.01). With regard to the echocardiographic findings, no differences were found with regards to left ventricular morphology, neither systolic nor diastolic function (LVEF 32.3 ± 6.9 and 32.1 ± 7.3%; left ventricular end-diastolic diameter 63.5 ± 8.5 and 62.3 ± 8.5 mm; left ventricular end-diastolic volume indexed 97.2 ± 33.2 and 96.3 ± 34.3 mL/m², in MHDS and NO-MHDS, respectively).

With regard to changes in peak VO₂ from baseline during follow-up, available data (about 20% of final cohort) showed that MHDS patients displayed a more severe impairment when compared with NO-MHDS patients (data expressed as delta changes from baseline: -4.5 ± 0.3 vs. -2.1 ± 0.2 respectively, *P* = 0.001).

Discussion

In the present study, the first prospective registry specifically designed to investigate the role played by HD and diabetes on prognosis and survival of heart failure patient with ejection fraction ≤45%, we document several novel findings: (i) the prevalence of HD in CHF is very high, exceeding 90%. Specifically, more than two-thirds of patients are characterized by two or more concomitant HD, i.e. multiple hormone and metabolic deficiency syndrome (MHDS); (ii) the presence of MHDS identifies a group of patients at increased risk of mortality or cardiovascular hospitalization; (iii) there is a graded relation between the number of HD and total events, suggesting a possible causal role of MHDS in CHF; and (iv) T2D, but not IR, exerts a negative role on CHF survival and prognosis.

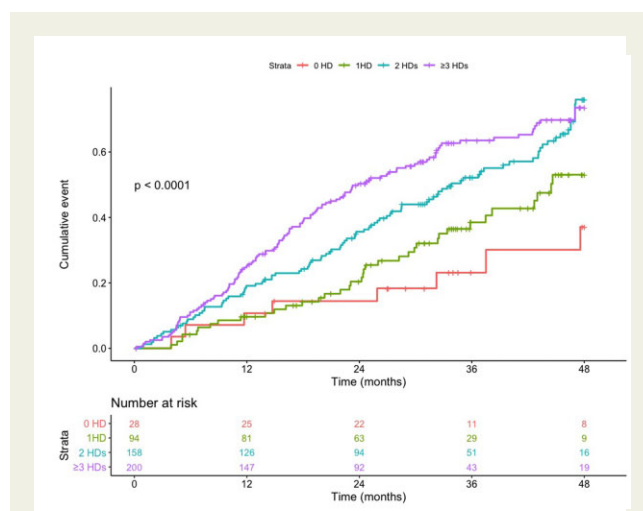


Figure 3 Occurrence of the primary endpoint according to the number of hormonal deficiencies. Kaplan–Meier analysis of survival for all-cause mortality or cardiovascular hospitalization in patients with 0 ($n = 28$), 1 ($n = 94$), 2 ($n = 158$), three, or more than 4 ($n = 200$) hormonal deficiencies [1.35 (1.21–1.52), $P < 0.001$]. Log-rank testing was applied for calculation of P -values. MHDS, multiple hormonal and metabolic deficiencies syndrome.

Hormone deficiencies and heart failure

Recently, the new concept that CHF progression is associated with both the overactivity of adrenergic/renin–angiotensin–aldosterone/cytokine systems and the down-regulation/impairment of hormone activities has emerged.⁵ Such a pathophysiological model stems from several independent studies documenting that isolated HD (i.e. adrenal, somatotrophic, gonadal, thyroid, and insulin axes) were associated with the impairment of clinical status and physical performance, and with poor survival.^{6,10,11,14–16} However, data available in the literature are scattered, lacking an overall view of the phenomenon. In addition, even when the effects of the combination of HDs have been studied in smaller cohorts, IR or diabetes have been disregarded.⁶ The T.O.S.C.A. registry overcomes this limitation, adding valuable information in the topic, in view of the even more emerging role of IR or diabetes as relevant players in the pathogenesis of CHF.¹²

The T.O.S.C.A. registry shows that more than 90% of the patients with CHF have at least one HD. Consistently with previous results, the presence of TD, DHEAS-D, or IGF-1 deficiency were independently associated with poor outcome. Specifically, the role played by each component of the somatotrophic axis (IGF-1 and GH) has been extensively studied during the last 20 years, with several independent groups reporting lower circulating IGF-1 levels associated with worst NYHA class, impaired exercise capacity, sarcopenia, and increased inflammatory activation.^{17–20} GH deficiency is also associated with impaired clinical status, left ventricular remodelling, right ventricular dysfunction, and increased mortality.¹⁴ Likewise, TD is associated with significant impairment of skeletal muscle function and exercise capacity,^{21–23} as well as a dysregulation of metabolic profiles, with increased risk of metabolic syndrome and new onset of T2D.²⁴ Similarly, low T3 syndrome has been associated with worse

cardiovascular performance and increased mortality in CHF,¹⁵ and restoring circulating thyroid hormone levels in low T3 syndrome CHF patients has been shown to improve cardiac output and induce neurohormonal deactivation.²⁵ IR and T2D have been associated with the severity of CHF, as testified by impaired 6-min walking test and reduced peak VO_2 .^{10,25,26}

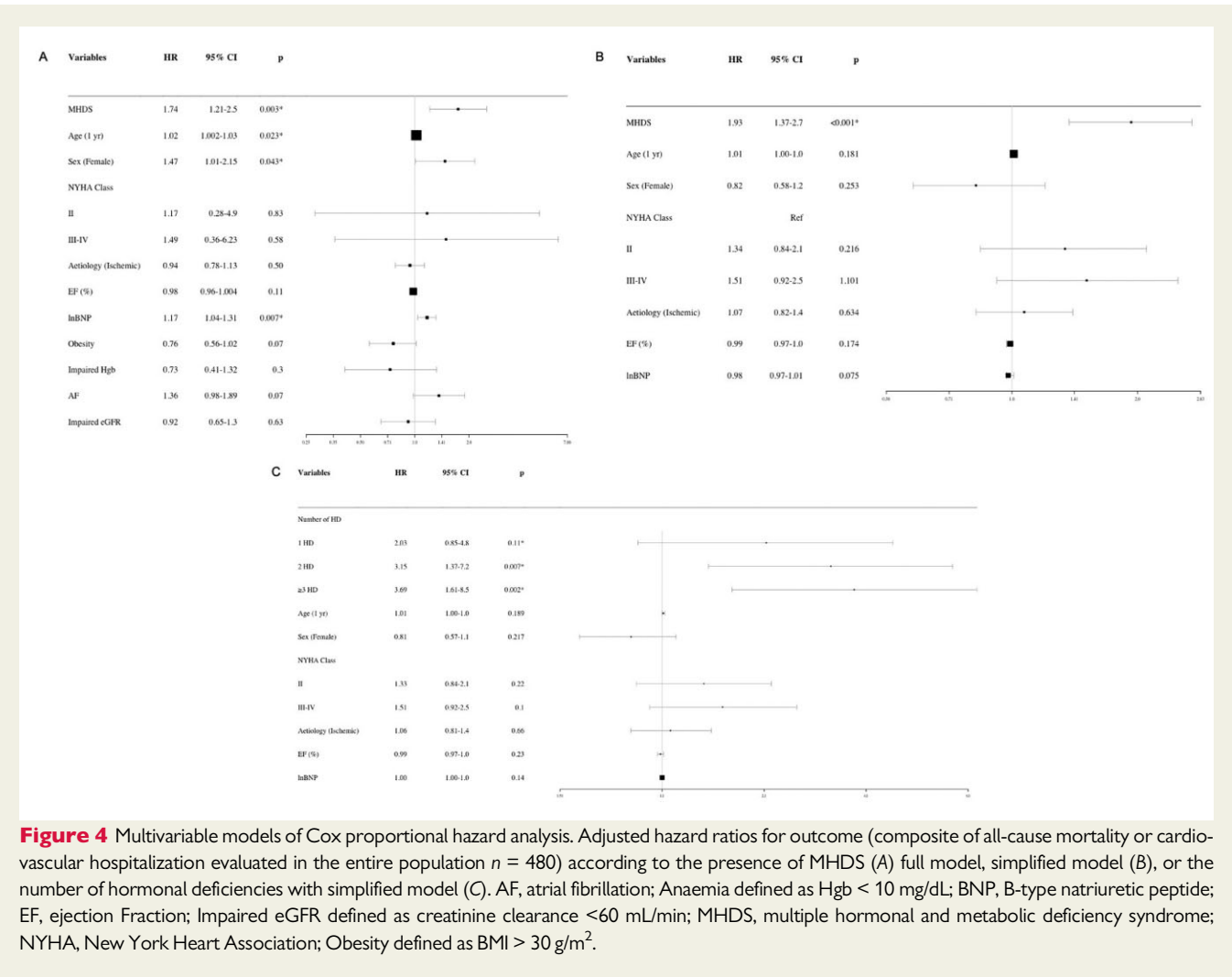
In the current study, we initially combined in a unique group the patients with T2D and the patients without T2D but with IR. This group, about two-thirds of the entire population, was characterized by an increased occurrence of the primary outcome. T2D is a complex syndrome in which IR and hyperglycaemia coexist and weighting the relative role played by each of these two components is complex. However, when T2D alone was investigated, it exerts a tremendous impact on outcome, even when adjusted in the multivariate analysis, confirming observations from literature.²⁷ On the other hand, when patients with IR without T2D were investigated, we did not find any effect of IR on the prognosis of heart failure. Our finding suggests that, at least on the background of the treatment currently in use, IR does not appear to play a relevant role in the progression of CHF. Intriguingly, it has been demonstrated that drugs acting on insulin resistance (i.e. glitazones), were non-effective in CHF, whereas drugs having no direct effect on insulin sensitivity (i.e. sodium-glucose co-transporter-2 inhibitors). Considering that almost 40% of our patients are IR but non-diabetics, to clarify the role played by IR *per se* is very important, and further analyses are needed.

Taken together, the T.O.S.C.A. registry confirmed that each of these HD in CHF is associated not only with a worse clinical status and impaired performance, but, more importantly, with reduced survival in CHF, further suggesting their causal role in CHF progression.^{10,11,13–18,27} Current data do not display that a single hormonal defect has a predominant effect on morbidity and mortality compared with the others: except low T3, the survival curves related to the individual hormones display a similar divergence over time, pointing to the concept that each HD bears similar effect on outcome, while their aggregation portends to worse prognosis. Indeed, according to the interaction analysis performed in our work, none of the HD was able to drive the primary outcome.

Multiple hormone and metabolic deficiency syndrome in heart failure

Our report showed that the coexistence of at least two HD (i.e. MHDS) is very common, involving about 75% of subjects; notably, MDHS is strongly associated with impaired cardiovascular performance and increased risk of hospitalization and death.

Previous preliminary findings suggested that the coexistence of HD have a high prevalence among the patients with CHF and might have an impact on its prognosis.^{6–8} Specifically, in a cohort of 208 male CHF patients, it has been demonstrated that the coexistence of at least two HD, among low levels of IGF-1, total testosterone, or DEHA-S (about 50% of the population) was associated with reduced survival.⁶ More recently, in 107 male patients, an MHDS has been described in about 30% of the population, with an association between the number of HD and the outcome.⁷ Finally, in a small cohort of 72 male patients, a prevalence of MHDS was found in about 60% of population, with a lower prevalence (about 40%) in heart failure with preserved ejection fraction.⁸



Going further these findings, we investigated a broader and more complete panel of hormones, showing also the impact of thyroid HD and IR/T2D. In the current report, we demonstrate, for the first time, that MHDS has an independent and heavy effect of mortality or cardiovascular hospitalization.

A further strength of the present report is that female patients represent about 20% of the investigated cohort, whereas in all previous studies on the topic they have always been excluded. Therefore, we provide the first demonstration of the importance of MHDS in a mixed-sex population. This is of utmost importance, given the low representations of women in clinical studies engaging heart failure patients and the unmet need to better characterize female patients affected by this clinical condition.

Finally, we demonstrated for the first time that MHDS is associated with a more dramatic progression of the disease, as testified by the marked change in VO₂ between MHDS and NO-MHDS from baseline to follow-up. Indeed, even if no differences were observed with regard to echocardiographic findings, MHDS was associated with a more prominent decline of peak oxygen consumption. This might be due to the loss of many protective effects of hormones on myocardial mechanics, left ventricular remodelling, but also to the recognized

action of anabolic hormones on skeletal muscle, endothelial function, and ventilator exchanges.⁵

Translational perspective

The demonstration that hormones play a pivotal role in CHF progression and predict survival opens novel therapeutic horizons. While most circulating biomarkers are accepted as molecular signatures capable of predicting relevant disease states or clinical outcomes or guiding therapy, HDs are endowed with the potential advantage of being correctable with appropriate replacement therapy, with preliminary positive results available in literature.^{23,28-33} Findings from the T.O.S.C.A. Registry suggest to searching for HD in patients with heart failure as promising therapeutic targets and support the need for clinical trials aimed to demonstrate the potential benefits of hormonal replacement therapy.

Limitations

The observational character of our study is acknowledged.¹³ Therefore, the study was not designed to elucidate the putative biological mechanism of HD in CHF. However, observational studies are more appropriate to describe the natural history of a disease and to

generate or confirm new pathophysiological hypothesis.³⁴ Furthermore, even if the cut-off values for defining the presence of HD may be regarded as somewhat arbitrary, they were chosen according with extant literature on the topic and, when available, with current guidelines.

Supplementary material

Supplementary material is available at *European Journal of Preventive Cardiology* online.

Acknowledgements

We are especially grateful to patients and nurses who have participated in this study. Dr Salzano receives research grant support from Cardiopath, Department of Advanced Biomedical Sciences, Federico II University of Naples, Naples, Italy, and UniNa and Compagnia di San Paolo, in the frame of the Programme STAR. Dr Marra was supported by an institutional grant from Italian Healthcare Ministry (Ricerca Finalizzata for young researchers) project GR-2016-02364727. The authors are grateful to Sekisui Medical Co. for provision of RapidPIA™ BNP kits. The T.O.S.C.A. Registry is an Investigator initiated trial with external funding being provided mainly by unrestricted grants from Merck Serono Italy.

Funding

The work was supported by unrestricted grant from Merck Serono Italy.

Conflict of interest: The authors declare that they have no conflict of interest.

Appendices

The T.O.S.C.A. investigators include: A. Cittadini, A.M. Marra, M. Arcopinto, R. D'Assante, L. Saccà, M.G. Monti, R. Napoli, M. Matarazzo, F.M. Stagnaro, L. Piccioli, A. Lombardi, V. Panicara, M. Flora, L. Golia, V. Faga, A. Ruocco, D. Della Polla, R. Franco, A. Schiavo, A. Gigante, E. Spina, M. Sicuranza, F. Monaco, M. Apicella, C. Miele, A.G. Campanino, L. Mazza, R. Abete, A. Farro, F. Luciano, R. Polizzi, G. Ferrillo, M. De Luca, G. Crisci, F. Giardino, M. Barbato (Department of Translational Medical Sciences, Federico II University, Naples, Italy); A. Salzano, B. Ranieri (IRCCS S.D.N., Naples, Italy); E. Bossone (AORN A Cardarelli, Naples, Italy); F. Ferrara, V. Russo, M. Malinconico, R. Citro (Heart Department, Cardiology Division, "Cava de' Tirreni and Amalfi Coast" Hospital, University of Salerno, Salerno, Italy); E. Guastamacchia, M. Iacoviello, M. Leone, (University of Bari "Aldo Moro", Bari, Italy); V. Triggiani, V.A. Giagulli (Interdisciplinary Department of Medicine-Section of Internal Medicine, Geriatrics, Endocrinology and Rare Diseases, University of Bari "A. Moro", Bari, Italy); F. Cacciatore, C. Maiello, C. Amarelli, I. Mattucci (Heart Transplantation Unit, Monaldi Hospital, Azienda Ospedaliera dei Colli, Naples, Italy); G. Limongelli, D. Masarone, P. Calabrò, R. Calabrò, A. D'Andrea, V. Maddaloni, G. Pacileo, R. Scarafile (Cardiology SUN, Monaldi Hospital, Azienda Ospedaliera dei Colli, Second University of Naples, Naples, Italy); F. Perticone, A. Belfiore, A. Sciacqua, A. Cimellaro (University Magna Graecia of Catanzaro, Catanzaro, Italy); P. Perrone Filardi, L. Casaretti, S. Paolillo, P.

Gargiulo (Department of Advanced Biomedical Sciences, Federico II University of Naples, Naples, Italy); A. Mancini, A.M.R. Favuzzi, C. Di Segni, C. Bruno, E. Vergani (Operative Unit of Endocrinology, Catholic University of the Sacred Heart, Rome); M. Volterrani, R. Massaro (IRCCS S. Raffaele Pisana, Roma, Italy); O. Vriz, F. Grimaldi (Azienda Ospedaliero-Universitaria "Santa Maria della Misericordia" San Daniele del Friuli, Udine, Italy); R. Castello, A. Frigo (Azienda Ospedaliera Universitaria Integrata di Verona, Italy); M.R. Campo, M.R. Sorrentino (Ospedali Riuniti di Foggia, Italy); P.A. Modesti, D. Malandrino (Università di Firenze, Italy); R. Manfredini, A. De Giorgi, F. Fabbian (Azienda Ospedaliera-Universitaria S. Anna, Ferrara, Italy); A. Puzzo, L. Ragusa (I.R.C.S.S. Oasi Maria SS, Troina, Italy); L. Caliendo, L. Carbone (Ospedale Santa Maria della Pietà, Nola, Napoli, Italy); A. Frigiola, T. Generali, F. Giacomazzi, C. De Vincentiis, A. Ballotta (IRCCS San Donato Milanese, Milano, Italy); P. Garofalo, G. Malizia (Ospedali Riuniti "Villa Sofia—Cervello", Palermo, Italy); S. Milano, G. Misiano (Policlinico P. Giaccone, Palermo, Italy); T. Suzuki, M.Z. Israr, D. Bernieh, S. Cassambai, Y. Yazaki (Department of Cardiovascular Sciences and NIHR Leicester Biomedical Research Centre, University of Leicester, Glenfield Hospital, Leicester, UK); L.M. Heaney (School of Sport, Exercise & Health Sciences, Loughborough University, Loughborough, UK); K.A. Eagle (Michigan Frankel Cardiovascular Center, University of Michigan, Ann Arbor, Michigan); H.O. Ventura (John Ochsner Heart and Vascular Institute, Ochsner Clinical School-the University of Queensland School of Medicine, New Orleans, LA, USA); A. Colao (Clinical Medicine and Surgery Department—Federico II University, Naples, Italy); D. Bruzzese, Statistical Management (Department of Public Health, University Federico II of Naples, Naples, Italy).

Data availability

The data that support the findings of this study are available from the corresponding author, upon reasonable request.

References

- Virani SS, Alonso A, Benjamin EJ, Bittencourt MS, Callaway CW, Carson AP, Chamberlain AM, Chang ALR, Cheng SS, Delling FN, Djousse L, Elkind MSV, Ferguson JF, Fornage M, Khan SS, Kissela BM, Knutson KL, Kwan TW, Lackland DT, Lewis TT, Lichtman JH, Longenecker CT, Loop MS, Lutsey PL, Martin SS, Matsushita K, Moran AE, Mussolino ME, Perak AM, Rosamond WD, Roth GA, Sampson UKA, Satou GM, Schroeder EB, Shah STH, Shay CM, Spartano NL, Stokes A, Tirschwell DL, VanWagner LB, Tsao CW; On behalf of the American Heart Association Council on Epidemiology and Prevention Statistics Committee and Stroke Statistics Subcommittee. Heart disease and stroke statistics-2020 update: a report from the American Heart Association. *Circulation* 2020;**141**:E139–E596.
- Meta-analysis Global Group in Chronic Heart Failure (MAGGIC). The survival of patients with heart failure with preserved or reduced left ventricular ejection fraction: an individual patient data meta-analysis. *Eur Heart J* 2012;**33**:1750–1757.
- Ponikowski P, Voors AA, Anker SD, Bueno H, Cleland JGF, Coats AJS, Falk V, Gonzalez-Juanatey JR, Harjola VP, Jankowska EA, Jessup M, Linde C, Nihoyannopoulos P, Parissis JT, Pieske B, Riley JP, Rosano GMC, Ruilope LM, Ruschitzka F, Rutten FH, van der Meer P; ESC Scientific Document Group. 2016 ESC Guidelines for the diagnosis and treatment of acute and chronic heart failure: the Task Force for the diagnosis and treatment of acute and chronic heart failure of the European Society of Cardiology (ESC) Developed with the special contribution of the Heart Failure Association (HFA) of the ESC. *Eur J Heart Fail* 2016;**18**:891–975.
- Yancy CW, Jessup M, Bozkurt B, Butler J, Casey DE, Colvin MM, Drazner MH, Filippatos GS, Fonarow GC, Givertz MM, Hollenberg SM, Lindenfeld J, Masoudi FA, McBride PE, Peterson PN, Stevenson LW, Westlake C. 2017 ACC/AHA/HFSA focused update of the 2013 ACCF/AHA Guideline for the management of heart failure: a report of the American College of Cardiology/American Heart

- Association Task Force on Clinical Practice Guidelines and the Heart Failure Society of America. *Circulation* 2017;**136**:e137–e161.
5. Sacca L. Heart failure as a multiple hormonal deficiency syndrome. *Circ Heart Fail* 2009;**2**:151–156.
 6. Jankowska EA, Biel B, Majda J, Szklarska A, Lopuszanska M, Medras M, Anker SD, Banasiak W, Poole-Wilson PA, Ponikowski P. Anabolic deficiency in men with chronic heart failure—prevalence and detrimental impact on survival. *Circulation* 2006;**114**:1829–1837.
 7. Arcopinto M, Salzano A, Bossone E, Ferrara F, Bobbio E, Sirico D, Vriz O, De Vincentiis C, Matarazzo M, Saldamarco L, Saccà F, Napoli R, Iacoviello M, Triggiani V, Isidori AM, Vigorito C, Isgaard J, Cittadini A. Multiple hormone deficiencies in chronic heart failure. *Int J Cardiol* 2015;**184**:421–423.
 8. Salzano A, Marra AM, Ferrara F, Arcopinto M, Bobbio E, Valente P, Polizzi R, De Vincentiis C, Matarazzo M, Saldamarco L, Saccà F, Napoli R, Monti MG, D'Assante R, Isidori AM, Isgaard J, Ferrara N, Filardi PP, Perticone F, Vigorito C, Bossone E, Cittadini A. Multiple hormone deficiency syndrome in heart failure with preserved ejection fraction. *Int J Cardiol* 2016;**225**:1–3.
 9. Marra AM, Arcopinto M, Salzano A, Bobbio E, Milano S, Misiano G, Ferrara F, Vriz O, Napoli R, Triggiani V, Perrone-Filardi P, Sacca F, Giallauria F, Isidori AM, Vigorito C, Bossone E, Cittadini A. Detectable interleukin-9 plasma levels are associated with impaired cardiopulmonary functional capacity and all-cause mortality in patients with chronic heart failure. *Int J Cardiol* 2016;**209**:114–117.
 10. Suskin N, McKelvie RS, Burns RJ, Latini R, Pericak D, Probstfield J, Rouleau JL, Sigouin C, Solymoss CB, Tsuyuki R, White M, Yusuf S. Glucose and insulin abnormalities relate to functional capacity in patients with congestive heart failure. *Eur Heart J* 2000;**21**:1368–1375.
 11. Doehner W, Rauchhaus M, Ponikowski P, Godtsland IF, von Haehling S, Okonko DO, Leyva F, Proudler AJ, Coats AJS, Anker SD. Impaired insulin sensitivity as an independent risk factor for mortality in patients with stable chronic heart failure. *J Am Coll Cardiol* 2005;**46**:1019–1026.
 12. McMurray JJV, Solomon SD, Inzucchi SE, Køber L, Kosiborod MN, Martinez FA, Ponikowski P, Sabatine MS, Anand IS, Bělohávek J, Böhm M, Chiang C-E, Chopra VK, de Boer RA, Desai AS, Diez M, Drozd J, Dukát A, Ge J, Howlett JG, Katova T, Kitakaze M, Ljungerman CEA, Merkely B, Nicolau JC, O'Meara E, Petrie MC, Vinh PN, Schou M, Tereshchenko S, Verma S, Held C, DeMets DL, Docherty KF, Jhund PS, Bengtsson O, Sjöstrand M, Langkilde AM; DAPA-HF Trial Committees and Investigators. Dapagliflozin in patients with heart failure and reduced ejection fraction. *N Engl J Med* 2019;**381**:1995–2008.
 13. Bossone E, Arcopinto M, Iacoviello M, Triggiani V, Cacciatore F, Maiello C, Limongelli G, Masarone D, Perticone F, Sciacqua A, Perrone-Filardi P, Mancini A, Volterrani M, Vriz O, Castello R, Passantino A, Campo M, Modesti PA, De Giorgi A, Monte I, Puzzo A, Ballotta A, Caliendo L, D'Assante R, Marra AM, Salzano A, Suzuki T, Cittadini A; TOSCA Investigators. Multiple hormonal and metabolic deficiency syndrome in chronic heart failure: rationale, design, and demographic characteristics of the T.O.S.C.A. Registry. *Intern Emerg Med* 2018;**13**:661–671.
 14. Arcopinto M, Salzano A, Giallauria F, Bossone E, Isgaard J, Marra AM, Bobbio E, Vriz O, Åberg DN, Masarone D, De Paulis A, Saldamarco L, Vigorito C, Formisano P, Niola M, Perticone F, Bonaduce D, Saccà L, Colao A, Cittadini A; T.O.S.C.A. (Trattamento Ormonale Scompenso CArdiaco) Investigators. Growth hormone deficiency is associated with worse cardiac function, physical performance, and outcome in chronic heart failure: insights from the T.O.S.C.A. GHD Study. *PLoS One* 2017;**12**:e0170058.
 15. Iervasi G, Pingitore A, Landi P, Raciti M, Ripoli A, Scarlattini M, L'Abbate A, Donato L. Low-T3 syndrome—a strong prognostic predictor of death in patients with heart disease. *Circulation* 2003;**107**:708–713.
 16. Jankowska EA, Rozentryt P, Ponikowska B, Hartmann O, Kustrzycka-Kratochwil D, Reczuch K, Nowak J, Borodulin-Nadzieja L, Polonski L, Banasiak W, Poole-Wilson PA, Anker SD, Ponikowski P. Circulating estradiol and mortality in men with systolic chronic heart failure. *JAMA* 2009;**301**:1892–1901.
 17. Niebauer J, Pflaum CD, Clark AL, Strasburger CJ, Hooper J, Poole-Wilson PA, Coats AJS, Anker SD. Deficient insulin-like growth factor I in chronic heart failure predicts altered body composition, anabolic deficiency, cytokine and neurohormonal activation. *J Am Coll Cardiol* 1998;**32**:393–397.
 18. Anker SD, Volterrani M, Pflaum CD, Strasburger CJ, Osterziel KJ, Doehner W, Ranke MB, Poole-Wilson PA, Giustina A, Dietz R, Coats AJS. Acquired growth hormone resistance in patients with chronic heart failure: Implications for therapy with growth hormone. *J Am Coll Cardiol* 2001;**38**:443–452.
 19. D'Assante R, Napoli R, Salzano A, Pozza C, Marra AM, Arcopinto M, Perruolo G, Milano S, Formisano P, Saldamarco L, Cirillo P, Cittadini A. Human heart shifts from IGF-1 production to utilization with chronic heart failure. *Endocrine* 2019;**65**:714–716.
 20. D'Assante R, Arcopinto M, Rengo G, Salzano A, Walser M, Gambino G, Monti M, Bencivenga L, Marra A, Åberg D, De VC, Ballotta A, Bossone E, Isgaard J, Cittadini A. Myocardial expression of somatotrophic axis, adrenergic, and calcium handling genes in heart failure with preserved ejection fraction and heart failure with reduced ejection fraction. *ESC Heart Fail* 2021; doi: 10.1002/ehf2.13067.
 21. Pugh PJ, Jones RD, West JN, Jones TH, Channer KS. Testosterone treatment for men with chronic heart failure. *Heart* 2004;**90**:446–447.
 22. Malkin CJ, Pugh PJ, West JN, van Beek EJR, Jones TH, Channer KS. Testosterone therapy in men with moderate severity heart failure: a double-blind randomized placebo controlled trial. *Eur Heart J* 2006;**27**:57–64.
 23. Caminiti G, Volterrani M, Iellamo F, Marazzi G, Massaro R, Miceli M, Mammi C, Piepoli M, Fini M, Rosano GMC. Effect of long-acting testosterone treatment on functional exercise capacity, skeletal muscle performance, insulin resistance, and baroreflex sensitivity in elderly patients with chronic heart failure: a double-blind, placebo-controlled, randomized study. *J Am Coll Cardiol* 2009;**54**:919–927.
 24. Oh JY, Barrett-Connor E, Wedick NM, Wingard DL. Endogenous sex hormones and the development of type 2 diabetes in older men and women: the Rancho Bernardo Study. *Diabetes Care* 2002;**25**:55–60.
 25. Pingitore A, Galli E, Barison A, Iervasi A, Scarlattini M, Nucci D, L'Abbate A, Mariotti R, Iervasi G. The acute effects of triiodothyronine replacement therapy in patients with chronic heart failure and low T3 syndrome: a randomised, placebo-controlled study. *Eur Heart J* 2007;**28**:648–648.
 26. Swan JW, Anker SD, Walton C, Godtsland IF, Clark AL, Leyva F, Stevenson JC, Coats AJS. Insulin resistance in chronic heart failure: Relation to severity and etiology of heart failure. *J Am Coll Cardiol* 1997;**30**:527–532.
 27. Chandramouli C, Teng T-HK, Tay WT, Yap J, MacDonald MR, Tromp J, Yan L, Siswanto B, Reyes EB, Ngarmukos T, Yu C-M, Hung C-L, Anand I, Richards AM, Ling LH, Regensteiner JG, Lam CSP, Mark Richards A, Lam CSP, Anand I, Hung C-L, Ling LH, Liew HB, Narasimhan C, Ngarmukos T, Park SW, Reyes E, Siswanto BB, Shimizu W, Zhang S; on behalf of the ASIAN-HF Investigators. Impact of diabetes and sex in heart failure with reduced ejection fraction patients from the ASIAN-HF registry. *Eur J Heart Fail* 2019;**21**:297–307.
 28. Arcopinto M, Bobbio E, Bossone E, Perrone-Filardi P, Napoli R, Saccà L, Cittadini A. The GH/IGF-1 axis in chronic heart failure. *Endocr Metab Immune Disord Drug Targets* 2013;**13**:76–91. doi: 10.2174/1871530311313010010.
 29. Cittadini A, Saldamarco L, Marra AM, Arcopinto M, Carlomagno G, Imbriaco M, Del Forno D, Vigorito C, Merola B, Oliviero U, Fazio S, Sacca L. Growth hormone deficiency in patients with chronic heart failure and beneficial effects of its correction. *J Clin Endocrinol Metab* 2009;**94**:3329–3336.
 30. Cittadini A, Marra AM, Arcopinto M, Bobbio E, Salzano A, Sirico D, Napoli R, Colao A, Longobardi S, Baliga RR, Bossone E, Sacca L. Growth hormone replacement delays the progression of chronic heart failure combined with growth hormone deficiency: an extension of a randomized controlled single-blind study. *JACC Heart Fail* 2013;**1**:325–330.
 31. Salzano A, Marra AM, Arcopinto M, D'Assante R, Triggiani V, Coscioni E, Pasquali D, Rengo G, Suzuki T, Bossone E, Cittadini A. Combined effects of growth hormone and testosterone replacement treatment in heart failure. *ESC Heart Fail* 2019;**6**:1216–1221.
 32. Salzano A, Marra AM, D'Assante R, Arcopinto M, Suzuki T, Bossone E, Cittadini A. Growth hormone therapy in heart failure. *Heart Fail Clin* 2018;**14**:501–515.
 33. Salzano A, Marra AM, Arcopinto M, D'Assante R, Triggiani V, Coscioni E, Pasquali D, Rengo G, Suzuki T, Bossone E, Cittadini A. Combined effects of growth hormone and testosterone replacement treatment in heart failure. *ESC Heart Fail* 2019;**6**:1216–1221.
 34. Salzano A, Suzuki T, Squire IB, Cittadini A. Are heart failure observational studies still useful? 'No need to argue'. *Eur J Prev Cardiol* 2020;**4**:2047487320932258.