Urocortin-2 in Acute Heart Failure: Role as a Marker of Volume Overload and Pulmonary Hypertension

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Abstract: Urocortin (Ucn)-2 has shown promising therapeutic effects on heart failure (HF). However, there are still significant knowledge gaps regarding the role and modulation of the endogenous Ucn-2 axis in the cardiovascular system and, specifically, in acute HF. We evaluated Ucn-2 levels in admission serum samples of 80 acute HF patients and assessed their association with clinical, analytical and echocardiographic parameters. Median age was 76.5 years, and 37 patients (46%) were male. Median serum Ucn-2 was 2.3ng/mL. Ucn-2 levels were positively associated

[1] Both authors contributed equally to the study.

All the authors take responsibility for all aspects of the reliability and freedom from bias of the data presented and their discussed interpretation.

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with peripheral edemas ($P = 0.022$), hepatomegaly ($P = 0.007$) and sodium retention score ($\rho = 0.37$, $P = 0.001$) and inversely correlated with inferior vena cava collapse at inspiration ($\rho = -0.37$, $P = 0.001$). Additionally, patients with higher Ucn-2 levels had a higher prevalence of right atrial dilation ($P = 0.027$), right ventricle dilation ($P = 0.008$), and higher systolic pulmonary artery pressure ($\rho = 0.34$, $P = 0.002$). Regarding analytical parameters, Ucn-2 correlated positively with log BNP ($r = 0.22$, $P = 0.055$) and inversely with uric acid ($r = 0.24$, $P = 0.029$) and total ($r = -0.30$, $P = 0.007$) and low-density lipoprotein cholesterol ($r = -0.23$, $P = 0.038$). No associations were found between Ucn-2 and age, sex or left heart structure or function. In conclusion, Circulating Ucn-2 was associated with clinical and echocardiographic markers of volume overload and pulmonary hypertension in acute HF patients. (Curr Probl Cardiol 2022;47:100860.)

Introduction

Acute heart failure (HF) is broadly defined as a rapid onset of new or worsening signs and symptoms of HF,[1] contributing largely to HF's worldwide burden. Acute HF is associated with high mortality, poor prognosis, and a high rehospitalization rate.[1]

Urocortin-2 (Ucn-2) is an endogenous vasoactive peptide of the corticotropin-releasing hormone (CRH) family that binds with high affinity to the type 2 CRH receptor (CRHR2), which is abundantly expressed in the cardiovascular system.[2] Ucn-2 promotes important hemodynamic effects, including vasodilation and positive cardiac inotropic, chronotropic, and lusitropic actions.[3,4] Additionally, the Ucn-2/CRHR2 system exhibits cardioprotective actions, preventing cell necrosis and apoptosis[5,6] and decreasing infarct size after ischemia-reperfusion injury in the rat heart.[6-8] Various experimental and human studies on HF have shown a potential therapeutic benefit upon Ucn-2 administration.[9-13] In an ovine model of pacing-induced HF, Ucn-2 administration synergized with conventional HF therapies, including angiotensin-converting-enzyme inhibitors (ACEi), diuretics, beta-adrenergic receptor blockers, and mineralocorticoid receptor antagonists (MRA), potentiating their benefits and blunting some of their adverse events.[14-17] Also, in patients with acute[13] and chronic HF,[12,18]

Ucn-2 administration resulted in a clearly augmented cardiac output without significant reflex tachycardia or systemic vasodilation.

Coupled with the promising findings regarding HF treatment, interest has grown in the potential role of Ucn-2 as a circulating marker of heart disease. Studies have described elevated Ucn-2 circulating levels in patients with coronary artery disease and left ventricular (LV) systolic dysfunction[19] and in non-ischemic dilated cardiomyopathy.[20] Additionally, Liew et al reported an inverse association between N-terminal-proUcn-2 (NT-proUcn-2) plasma levels and 2-years survival in HF.[21]

More recently, an association between Ucn-2 and pulmonary vasculature modulation and right ventricular (RV) function has been suggested.[22] Indeed, our group recently described an increased expression of Ucn-2 and CRHR2 in the RV of patients with pulmonary arterial hypertension (PAH) and a beneficial effect of Ucn-2 treatment in a rat model of monocrotaline (MCT)-induced pulmonary hypertension (PH).[22]

Despite the significant advances in our understanding of the Ucn-2 physiological roles in recent years and the encouraging results regarding its clinical use in HF treatment, there are still significant knowledge gaps regarding the role and modulation of the endogenous Ucn-2/CRHR2 axis in the cardiovascular system and, specifically, in acute HF. This study aimed to evaluate Ucn-2 circulating levels in a population of acute HF patients and their association with clinical and echocardiographic parameters of left and right heart structure and function.

Methods

Study population

The study population was part of a registry of acute HF conducted at the Internal Medicine Department of São João Hospital Centre, Porto, Portugal. Patients admitted with the primary diagnosis of acute HF were eligible for registry inclusion; patients with acute coronary syndromes were excluded. All patients underwent detailed and standardized clinical history, physical examination, and venous blood sample analysis at admission. An echocardiogram was performed within 72 hours after admission. Informed consent was obtained from each patient. The study protocol conforms to the ethical guidelines of the 1975 Declaration of Helsinki as reflected in a priori approval by the institution's human research committee.

From this registry, 80 acute HF patients were analyzed. We defined the following exclusion criteria: (1) HF due to severe valve disease; (2) rare

causes of HF (such as active myocarditis, congenital heart disease, or pericardial disease); (3) acute coronary syndrome in the past 3 months; and (4) severe chronic kidney disease, defined as a glomerular filtration rate <15 mL/min/1.73m^2 estimated by the MDRD formula.

Patients' admission volemia status was characterized using sodium retention score as previously described.[23] This score includes physical examination findings such as third sound gallop, rales, jugular venous distention, hepatomegaly, peripheral oedemas, and weight change, with higher values suggesting higher volume overload.

Echocardiographic evaluation

Images were obtained with a standard ultrasound machine (Vivid S6; GE Vingmed, Horten, Norway) using a multi-frequency matrix probe (2.0-3.6MHz). Standard techniques were used to obtain M-mode, 2D, and Doppler measurements following the European Association of Cardiovascular Imaging and the American Society of Echocardiography guidelines.

Left ventricle ejection fraction (LVEF), end-systolic and end-diastolic volumes were calculated by the biplane Simpson's method from apical 4- and 2-chamber views. Left atrial volume was calculated with the biplane disk summation technique. E/E' ratio was calculated from the relation of the E atrial wave measured by pulsed wave transmitral Doppler to the early diastolic velocity E' (average from the medial and lateral mitral annular E') by pulsed-wave tissue Doppler imaging. RV systolic function was evaluated by the tricuspid annular plane systolic excursion (TAPSE); RV systolic dysfunction was defined by a TAPSE<17 mm. Right atrial (RA) dimensions were estimated from an apical 4-chamber view using the disk summation technique; RA dilation was defined by RA volume indexes >32mL/m^2 in men and >27mL/m^2 in women.[24] RV dimensions were estimated at end-diastole from a RV−focused apical 4-chamber view; RV dilation was defined as a RV diameter >41 mm at the base and >35 mm at the mid-level.[24] Systolic pulmonary artery pressure (sPAP) was estimated from peak tricuspid regurgitation (TR) velocity, using the simplified Bernoulli equation.[25] In 9 patients, TR velocity was not measurable and there were no signs of PH; in these patients, sPAP was set at 25 mm Hg for data analysis.

Samples collection and biomarkers measurement

Fasting venous blood samples were collected to a serum separator tube from Venosafe within the first 48 hours of hospital admission. Blood was

allowed to clot for 30 minutes and then centrifuged at 4500 rpm for 15 minutes at 20˚C. The serum was immediately separated and stored at -80˚C.

Serum biochemical parameters were measured using conventional methods with an Olympus AU5400 automated clinical chemistry analyzer (Beckman-Coulter, Izasa, Porto, Portugal). Plasma B-type natriuretic peptide (BNP) was measured using an Architect i2000 automated analyzer (Abbott, Lisbon, Portugal).

Ucn-2 serum levels were quantified in admission samples using an enzyme-linked immunosorbent assay (ELISA) kit (Product no. SEC585Hu, Cloud-Clone Corp, Wuhan, China), with a detection range of 0.78-50 ng/mL. The minimum detectable dose of this kit is typically less than 0.29 ng/mL. This assay has high sensitivity with an excellent specificity for the detection of Ucn-2 and has no significant cross-reactivity or interference with Ucn-2 analogs. The Ucn-2 level was measured according to the manufacturer's protocol and samples were run in duplicate. Optic density values were analyzed according to the values of standards.

Statistical analysis

For continuous variables, normality was assessed by the Shapiro-Wilks test, combined with a visual inspection of the histograms. If normally distributed, the results were summarized by the mean and standard deviation; otherwise, the median and the interquartile range (IQR) were used. Absolute (n) and relative (%) frequencies were reported for the categorical variables.

Univariate statistical analysis was carried out to assess the association between clinical variables and serum Ucn-2 levels and its effect-size.[26]

Multivariable-adjusted ordinary least squares regression was used to examine the relationship between Ucn-2 serum levels and variables most revealing volume and right heart overloads. The covariates were chosen based on the potential association found in Table 2 and in reports in the literature. Partial effects plots were used to reveal the link between the selected variables and the Ucn-2 serum levels.[27]

Results

Table 1 presents acute HF patients' characteristics at admission. Median age was 76 (IQR: 67, 82) years, and 37 patients (46%) were male. Thirty-eight patients (48%) had preserved ejection fraction (LVEF≥50%). The most frequent etiology was ischemic heart disease (46.2%). Median serum Ucn-2 at admission was 2.30 (IQR: 1.72, 3.40)

Table 1. Acute heart failure patients' characteristics at admission.

Characteristic*	N	N = 80
Patients' characteristics		
Sex	80	
Female		43 (54%)
Male		37 (46%)
Age (years)	80	76 (67, 82)
Atrial fibrillation	77	39 (51%)
Hypertension history	79	56 (71%)
Diabetes mellitus	80	31 (39%)
Body mass index (kg/m^2)	77	25.5 ± 5.1
Smoking history	80	
Never smoke		48 (60%)
Former smoker		26 (32%)
Current smoker		6 (7.5%)
Medication at admission		
ACEi or ARB	80	48 (60%)
Beta blocker	80	42 (52%)
Spirolactone	80	20 (25%)
Loop diuretic	80	63 (79%)
Physical examination at admission		
Systolic blood pressure (mm Hg)	76	120 ± 26
Diastolic blood pressure (mm Hg)	76	67 ± 14
Heart rate (bpm)	76	80 ± 16
Rales	79	
No Rales		19 (24%)
Lower one-third of the lungs		34 (43%)
Lower two-thirds of the lungs		21 (27%)
Throughout lung		5 (6.3%)
Jugular venous distension	75	22 (29%)
Hepatomegaly	73	14 (19%)
Peripheral oedemas	79	
No edema		17 (22%)
Leg		35 (44%)
Above leg		12 (15%)
Ankle		15 (19%)
Sodium retention score	70	4 (3, 5)
Echocardiographic evaluation		
LV ejection fraction ($\geq$ 50%)	79	38 (48%)
LAVI (mL/m^2)	76	52 ± 19
LV EDV (mL/m^2)	65	85 (51, 105)
LVMi (g/m^2)	76	131 ± 42
E/E'	75	19 ± 7
sPAP (mm Hg)	79	44 (35, 54)
RV systolic dysfunction	78	25 (32%)
RAVI (mL/m^2)	76	32 (24, 49)
RA dilation	76	43 (57%)
RV dilation	80	25 (31%)
IVC diameter (cm)	74	2.17 ± 0.56
IVC collapse at inspiration	72	38 (21, 58)

(*continued*)

Table 1. (*continued*)

Characteristic*	N	N = 80
IVC collapse at inspiration (≥50%)	72	30 (42%)
Admission laboratory parameters		
BNP (pg/mL)	77	1368 (646, 2717)
Natural log (BNP) (pg/mL)	77	7.22 (6.47, 7.91)
Hemoglobin (g/dL)	80	11.97 ± 1.94
Sodium (mEq/L)	80	138.0 ± 4.7
Uric acid (mg/L)	80	79 ± 28
Creatinine (mg/dL)	80	1.25 (1.05, 1.74)
GFR (MDRD) (mL/min)	80	49 (35, 63)
C-reactive protein (mg/L)	78	25 (11, 80)
Albumin (g/L)	80	35.2 (32.1, 38.4)
Total cholesterol (mg/dL)	80	154 ± 50
LDL cholesterol (mg/dL)	80	97 ± 37
Troponin I (ng/mL)	80	0.04 (0.02, 0.13)
Ucn-2 (pmol/L)	80	2.30 (1.72, 3.40)
Natural log (Ucn-2) (pmol/L)	80	0.83 ± 0.47

ACEi, angiotensin-converting enzyme inhibitor; ARB, angiotensin II receptor blocker; BNP, B-type natriuretic peptide; E/E' ratio, relation of the E atrial wave measured by pulsed wave transmitral Doppler and early diastolic velocity E' by pulsed-wave tissue Doppler imaging; GFR, glomerular filtration rate; IQR, interquartile range; IVC, inferior vena cava; LAVI, left atrial volume index; LDL, low-density lipoprotein; LV, left ventricle; LV EDV, left ventricle end-diastolic volume; LVMI, left ventricle mass index; MDRD, modification of diet in renal disease study; RA, right atria; RAVI, right atrial volume index; RV, right ventricle; sPAP, systolic pulmonary artery pressure; SD, standard deviation; Ucn-2, urocortin-2.

*Statistics presented: n(%); median (IQR); mean ± SD.

ng/mL. Ucn-2 levels were within the assay's detection range in every patient (0.9-7.0 ng/mL).

Table 2 summarizes the potential association between the measured clinical variables and the serum Ucn-2 levels. Table 2 reports no significant associations between Ucn-2 circulating levels and patients' age, sex, or previous comorbidities such as atrial fibrillation, hypertension, or diabetes *mellitus*.

Regarding physical examination findings, log Ucn-2 serum levels were not associated with pulmonary congestion (P= 0.117, Supplementary Fig 1) or with admission blood pressure. However, a significant positive association was found for peripheral edema (P= 0.022, Supplementary Fig. 2) and for the presence of hepatomegaly (P= 0.007). Overall, higher serum log Ucn-2 levels were associated with a significantly higher sodium retention score ($\rho = 0.37$, P = 0.001, Fig 1), suggesting higher volume overload. These findings were consistent with the echocardiographic data (Table 2), which showed an inverse

Table 2. Clinical association of serum Ucn-2 (natural log transformed) with clinical variables of enrolled acute heart failure patients.

Variable	N	Effect size	95% CI	*P* value
Patients' characteristics				
Age	80	-0.081*	[-0.29, 0.14]	0.460
Sex (female vs male)	80	-0.12‡	[-0.34, 0.10]	0.271
Atrial fibrillation	77	-0.08‡	[-0.33, 0.11]	0.469
Hypertension	79	0.01‡	[-0.20, 0.21]	0.961
Diabetes mellitus	80	-0.02‡	[-0.25, 0.19]	0.832
Physical examination at admission				
Systolic blood pressure	76	0.13†	[-0.10, 0.35	0.254
Diastolic blood pressure	76	0.19†	[-0.04, 0.40]	0.190
Rales	79	0.08§	[0.02, 0.20]	0.117
Peripheral edema	79	0.12§	[0.04, 0.30]	0.022
Hepatomegaly	73	0.31‡	[0.16, 0.48]	0.007
Sodium retention score	70	0.37*	[0.16, 0.62]	0.001
Echocardiographic evaluation				
LV ejection fraction (<50% vs ≥50%)	79	0.07‡	[-0.15, 0.30]	0.524
LAVI	76	0.10†	[-0.13, 0.32]	0.394
LV EDV	65	-0.03‡	[-0.27, 0.17]	0.804
LVMI	76	-0.19*	[-0.41, 0.02]	0.104
E/E'	75	-0.07*	[-0.30, 0.17]	0.550
sPAP	79	0.34*	[0.17, 0.54]	0.002
RV systolic dysfunction	78	-0.04‡	[-0.28, 0.19]	0.732
RA dilation	76	0.25‡	[0.06, 0.47]	0.027
RV dilation	80	0.30‡	[0.10, 0.51]	0.008
IVC collapse at inspiration	72	-0.37*	[-0.57, -0.16]	0.001
IVC diameter	74	0.19†	[-0.04, 0.40]	0.110
Admission laboratory parameters				
log BNP	77	0.22†	[-0.02, 0.44]	0.055
Hemoglobin	80	-0.02†	[-0.24, 0.20]	0.836
Sodium	80	-0.14†	[-0.35, 0.09]	0.227
Uric acid	80	0.24†	[0.03, 0.44]	0.029
Albumin	80	-0.07†	[-0.29, 0.15]	0.529
GFR (MDRD)	80	-0.10*	[-0.32, 0.10]	0.365
Total cholesterol	80	-0.30†	[-0.49, -0.09]	0.007
LDL cholesterol	80	-0.23†	[-0.43, -0.01]	0.038
Troponin I	80	0.12*	[-0.10, 0.33]	0.308

BNP, B-type natriuretic peptide; CI, confidence interval; E/E' ratio, relation of the E atrial wave measured by pulsed wave transmitral Doppler and early diastolic velocity E' by pulsed-wave tissue Doppler imaging; GFR, glomerular filtration rate; IVC, inferior vena cava; LAVI, left atrial volume index; LDL, low-density lipoprotein; LV, left ventricle; LV EDV, left ventricle end-diastolic volume; LVMI, left ventricle mass index; MDRD, Modification of Diet in Renal Disease Study; RA, right atria; RV, right ventricle; sPAP, systolic pulmonary artery pressure; Ucn-2, urocortin-2.

*Spearman correlation.

†Pearson correlation.

‡Mann-Whitney U-test.

§Kruskal-Wallis Rank sum test.

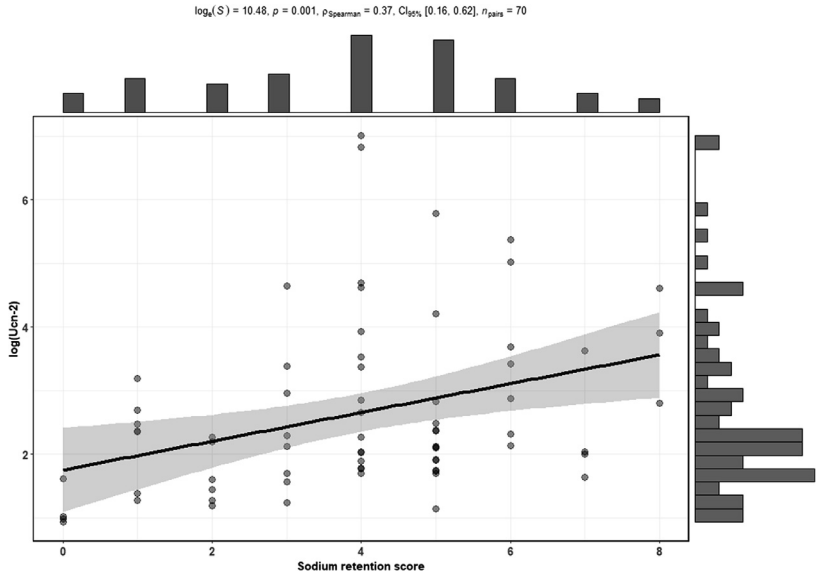

FIG 1. Scatter plot between Ucn-2 levels and sodium retention score.

correlation between Ucn-2 levels and inferior vena cava (IVC) collapse at inspiration ($\rho = -0.37$, $P = 0.001$, Supplementary Fig 3). Also, patients with higher log Ucn-2 levels had a higher prevalence of RA dilation ($p = 0.027$, Supplementary Fig 4), RV dilation ($P = 0.008$, Supplementary Fig 5) and higher sPAP ($\rho = 0.34$, $P = 0.002$, Supplementary Fig 6). No significant associations were found between log Ucn-2 levels and LVEF (Supplementary Fig 7), left chambers dimensions, LV mass, or E/E' ratio (Table 2). Regarding the association of log Ucn-2 levels with analytical parameters at admission, we observed a minor positive association with log BNP ($r = 0.22$, $P = 0.055$, Supplementary Fig 8). Also, Ucn-2 levels correlated positively with uric acid levels ($r = 0.24$, $P = 0.029$, Supplementary Fig 9) and inversely with total and low-density lipoprotein (LDL) cholesterol ($r = -0.30$, $P = 0.007$; $r = -0.23$, $P = 0.038$, respectively, Supplementary Fig 10). No association was observed between log Ucn-2 levels and hemoglobin, renal function, sodium, albumin, or troponin I.

Finally, we assessed the association of Ucn-2 with BNP and selected clinical variables most indicative of volume overload and right heart overload, such as sodium retention score, sPAP, RV dilation, RA dilation and IVC collapse at inspiration (Fig 2). Overall, Ucn-2 seems to be associated with these variables independently of BNP.

Discussion

In this study, we described, for the first time, a positive association between Ucn-2 circulating levels and clinical, analytical and echocardiographic markers of volume overload in acute HF patients. Namely, Ucn-2 was increased in patients with higher sodium retention score and correlated positively with BNP levels and negatively with IVC collapse at inspiration. Also, patients with higher Ucn-2 levels had a higher prevalence of RA and RV dilation and higher sPAP, suggesting an activation of the endogenous Ucn-2 system in states of higher volume overload and in PH and right heart overload. Interestingly, Ucn-2 was associated with volume overload and sPAP independently of BNP.

Ucn-2 is a peptide of the CRH family with an important role within the cardiovascular system, where it is prominently expressed along with its receptor CRHR2.[4,28] Its interesting hemodynamic, anti-hypertrophic and anti-ischemic effects led to the study of its therapeutic value in the setting of acute and chronic HF. In an ovine model of acute HF, Ucn-2 infusion led to increased cardiac contractility and output and arterial pressure

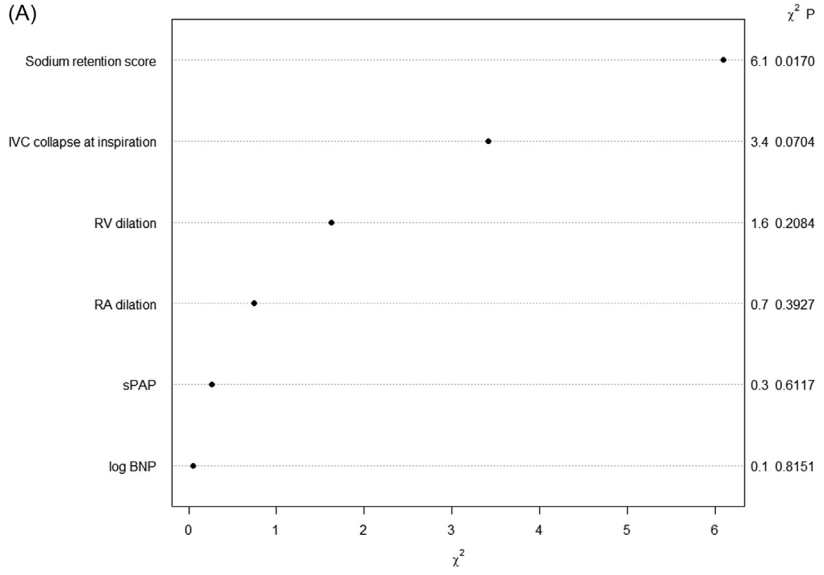

FIG 2. Associations of Ucn-2 levels in acute heart failure with a set of clinical variables; (A) Display of the relative contribution of each variable to Ucn-2 serum level, ranked according to their chi-square value, from the ordinary least square multivariable-adjusted model; and (B) Partial effects plot of the clinical variables linked to volume overload and right heart overload and the Ucn-2 serum levels.

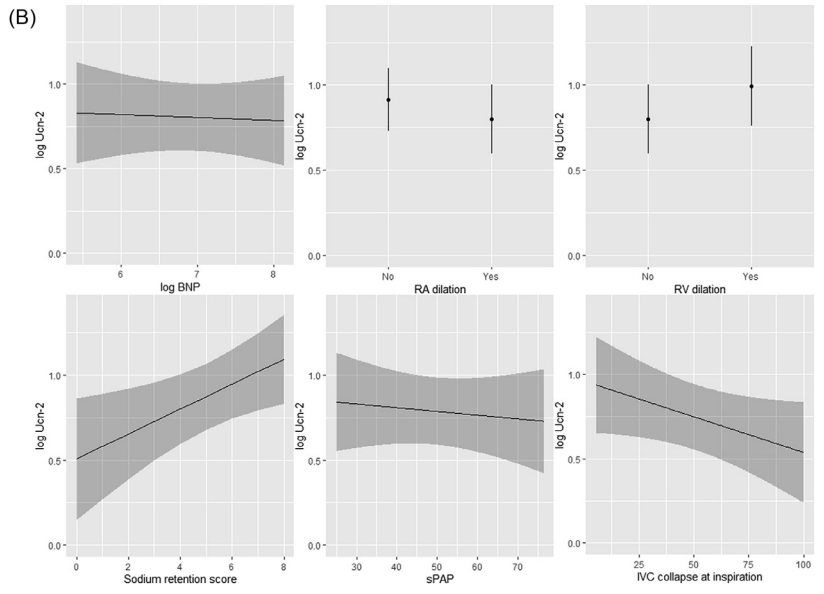

FIG 2 Continued.

similar to dobutamine infusion, with greater reductions in central venous and left atrial pressures.[9] In patients with acute HF, Ucn-2 infusion was administered as an adjunct to conventional therapy,[13] resulting in vasodilation, increased cardiac output without significant tachycardia and sustained fall in BNP levels. However, there was a transient worsening of renal function due to its hypotensive effect, and further investigation is necessary to better characterize Ucn-2 therapeutic potential.

Regarding the endogenous Ucn-2 system characterization, only a few reports address Ucn-2 circulating levels in cardiovascular diseases and specifically in HF,[12,19-21] with high variability between results complicating their comparison and interpretation. Davis et al found very low levels of Ucn-2 in healthy volunteers[29] and described baseline circulating levels of 230 (40-420) pg/mL in a small sample of patients with chronic stable HF with reduced EF (LVEF $\leq$ 40%).[12] Topal et al[19] studied patients with suspected coronary artery disease and compared Ucn-2 levels according to the degree of systolic dysfunction. They found increased Ucn-2 in patients with LVEF 40-55% when compared to controls (median 12.7pg/mL vs 11.0pg/mL, P = 0.047) but not in patients with LVEF$\leq$40% (median 8.9 pg/mL, P = 0.52). Curiously, patients with LVEF$\leq$40% had significantly lower Ucn-2 levels than patients with LVEF 40%-55% (P =

0.03). Tsuda et al[20] compared a group of stable non-ischemic dilated cardiomyopathy patients (LVEF<50%) to age and gender-matched healthy subjects and found increased plasma Ucn-2 levels in HF (median 1755 pg/mL vs 235 pg/mL, $P < 0.01$). Finally, Liew et al[21] found increased levels of NT-proUcn-2 in HF patients with preserved (119 [93-136] ng/L) or reduced EF (117 [98-141] ng/L) when compared to controls (112 [86-132] ng/L), and, interestingly, described an inverse association between NT-proUcn-2 levels and 2-year mortality in HF patients, independent of NT-proBNP, age and LVEF. This finding was intriguing, and the authors raise the hypothesis that patients with HF who were able to muster a more vigorous Ucn-2 response may have received some protection through that response.[21]

Our findings suggest activation of the endogenous Ucn-2 system as a counter-regulatory mechanism in states of volume overload, such as acute HF. Evidence in the literature supports this hypothesis. Namely, both animal and human studies on the administration of Ucn-2 in acute HF show a suppression of the classical deleterious neuro-hormonal systems, such as the renin-angiotensin-aldosterone system (RAAS),[9,13] arginine-vasopressin and sympathetic nervous system.[9] Furthermore, in a sheep model of HF, Ucn-2 use in combination with conventional HF therapies, including ACEi, diuretics, beta-adrenergic receptor blockers, and MRA, resulted in augmentation of their beneficial hemodynamic effects compared with either agent alone and in a reduction of their adverse effects.[14-17] A recent study also described increased Ucn-2 serum levels in patients with hypertension treated with ACEi,[30] suggesting that some beneficial RAAS blockade effects may be related to the Ucn-2 system.

We also evaluated the association between Ucn-2 levels and markers of right heart function and remodeling and pulmonary artery (PA) pressures and found that patients with higher Ucn-2 levels had more right chambers dilation and higher sPAP. Besides, Ucn-2 predicted parameters of volume overload and right heart overload independently of BNP, possibly reflecting a distinct activation pathway, more dependent on pulmonary and right heart pressures. Indeed, we found no association between Ucn-2 and left chambers dimensions or LV systolic or diastolic function, although the lack of a control group of patients with no HF may have precluded the identification of a correlation with these parameters. Studies on the role of Ucn-2 in the cardiovascular system focused mainly on the left heart function and the systemic vasculature, but recent evidence in the literature supports the association between Ucn-2, PH, and RV function.[31] Our group studied a sample of patients with PAH, with or without RV failure.[22] Although no differences were observed in plasma Ucn-2

levels between PAH and control patients, Ucn-2 levels were increased PAH patients' buffy-coat, suggesting an activation of the Ucn-2 endogenous system in circulating and inflammatory cells, known to mediate vascular and myocardial remodeling mechanisms.[22] Also, in PAH patients, Ucn-2 mRNA and CRHR2 protein levels were increased in the RV of patients with RV failure, and Ucn-2 mRNA levels correlated with the degree of RV dysfunction and mean PAP,[22] suggesting a compensatory up-regulation in response to increased myocardial stress, inflammation and vascular remodeling. Our group additionally studied a rat model of MCT-induced PH and found increased Ucn-2 plasma levels and Ucn-2 mRNA expression in the RV of MCT-injected rats[22] but not in the lung tissue of these animals, raising the hypothesis that Ucn-2 circulating levels may be more dependent on RV synthesis in response to myocardial overload than on lung synthesis. However, in a previous study on human HF, no significant increase in plasma Ucn-2 levels was found in coronary sinus blood when compared to peripheral arterial and venous blood,[20] so further studies are required to identify the cellular origin and the main stimuli for Ucn-2 secretion in this context. In rats with MCT-induced PH, chronic treatment with human Ucn-2 improved survival and exercise capacity, reduced pulmonary vascular resistance, and mitigated pulmonary RV remodeling and dysfunction.[22] Overall, these results strengthen our hypothesis of an association between Ucn-2 and right heart function and load. Thus, we may hypothesize that Ucn-2 secretion to the circulation in acute HF may be triggered by an increase in PA pressures and right heart load and may represent a counter-regulatory mechanism trying to overcome this increased load. Although the pilot study UNICORN could not demonstrate a clear effect of Ucn-2 infusion in the sPAP and RA pressure on patients with acute HF,[13] preclinical studies on a model of acute HF described a greater reduction in central venous pressure with Ucn-2 infusion than with dobutamine,[9] further supporting this hypothesis.

Finally, we found a positive association of Ucn-2 with uric acid levels and an inverse association with total and LDL cholesterol. To our knowledge, there are no previous reports regarding the association between urocortin and uric acid and only scarce data regarding its correlation with the metabolic profile. We may speculate that their correlation can be explained by HF severity, since lower cholesterol and higher uric acid levels are associated with states of more severe HF and worse outcomes. However, some reports recently suggested an inverse association between Ucn-2 and total and LDL cholesterol,[32,33] and additional evidence is needed to clarify these findings.

Study limitations

Our study has some limitations. The relatively small sample size may have hindered the identification of additional associations between Ucn-2 and other clinical, analytical and echocardiographic parameters. The cross-sectional design and the absence of a control group with no evidence of HF preclude the validation of Ucn-2 as a potential diagnosis and prognosis biomarker in acute HF and the establishment of a causal association between Ucn-2, volume overload and PH. Also, this study could not identify the origin and the targets of the circulating Ucn-2 detected, and additional studies are necessary to clarify the mechanisms of its synthesis, turnover and action in acute HF. Finally, it is important to note that Ucn-2 assays have not been internationally standardized and that there is little assay validation, which limits the comparison of Ucn-2 circulating levels between studies (which sometimes spread over a thousand-fold range), and the proper validation of Ucn-2 as a biomarker.[4,21] Differences in methodologies, such as different format assays and different epitopes targets of the antibodies used by the various assays, are likely to contribute to these inter-study variances. Nevertheless, although this technical issue may limit the external generalization of our findings regarding Ucn-2 absolute concentrations in HF patients' serum, we used the same batch of a commercially available assay for all Ucn-2 determinations, so it is unlikely that this methodological limitation affects the internal validation of our findings.

Conclusions

In conclusion, we describe, for the first time, an association between Ucn-2 and volume and right heart overload in acute HF, opening a new door to the comprehension of the role of the endogenous Ucn-2 system in HF. Further studies are required to better establish the role of Ucn-2 levels as a biomarker in this context and to explore Ucn-2 therapeutic potential in the setting of right heart overload and PH.

Supplementary materials

Supplementary material associated with this article can be found in the online version at doi:10.1016/j.cpcardiol.2021.100860.

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