

# Association of arginine vasopressin with low atrial natriuretic peptide levels, left ventricular remodelling, and outcomes in adults with and without heart failure

Julio A. Chirinos<sup>1,2,3\*†</sup>, Mayank Sardana<sup>4†</sup>, Garrett Oldland<sup>1,2</sup>, Bilal Ansari<sup>3</sup>, Jonathan Lee<sup>2</sup>, Anila Hussain<sup>3</sup>, Anique Mustafa<sup>3</sup>, Scott R. Akers<sup>1</sup>, Wen Wei<sup>5</sup>, Edward G. Lakatta<sup>5</sup> and Olga V. Fedorova<sup>5</sup>

<sup>1</sup>Corporal Michael J. Crescenz VAMC, Philadelphia, PA, USA; <sup>2</sup>University of Pennsylvania Perelman School of Medicine, Philadelphia, PA, USA; <sup>3</sup>Hospital of the University of Pennsylvania, Philadelphia, PA, USA; <sup>4</sup>University of Massachusetts Medical School, Worcester, MA, USA; <sup>5</sup>National Institute of Aging, Baltimore, MD, USA

## Abstract

**Aims** The arginine vasopressin (AVP) pathway has been extensively studied in heart failure (HF) with reduced ejection fraction (HFrEF), but less is known about AVP in HF with preserved EF (HFpEF). Furthermore, the association between AVP and atrial natriuretic peptide (ANP, a well-known inhibitor of AVP secretion) in HF is unknown.

**Methods and results** We studied subjects with HFpEF ( $n = 28$ ) and HFrEF ( $n = 25$ ) and without HF ( $n = 71$ ). Left ventricular (LV) mass and left atrial (LA) volumes were measured with cardiac magnetic resonance imaging. Arginine vasopressin and ANP were measured with enzyme-linked immunosorbent assay. Arginine vasopressin levels were significantly greater in HFpEF [0.96 pg/mL; 95% confidence interval (CI) = 0.83–1.1 pg/mL] compared with subjects without HF (0.69 pg/mL; 95% CI = 0.6–0.77 pg/mL;  $P = 0.0002$ ). Heart failure with preserved ejection fraction (but not HFrEF) was a significant predictor of higher AVP after adjustment for potential confounders. Arginine vasopressin levels were independently associated with a greater LA volume and also paradoxically, with lower ANP levels. Key independent correlates of higher AVP were the presence of HFpEF (standardized  $\beta = 0.32$ ; 95% CI = 0.09–0.56;  $P = 0.0073$ ) and the ANP/LA volume ratio (standardized  $\beta = -0.23$ ; 95% CI =  $-0.42$  to  $-0.04$ ;  $P = 0.0196$ ). Arginine vasopressin levels were independently associated with LV mass ( $\beta = 0.26$ ; 95% CI = 0.09–0.43;  $P = 0.003$ ) and with an increased risk of death or HF admissions during follow-up (hazard ratio = 1.61; 95% CI = 1.13–2.29;  $P = 0.008$ ).

**Conclusions** Arginine vasopressin is increased in HFpEF and is associated with LV hypertrophy and poor outcomes. Higher AVP is associated with the combination of LA enlargement and paradoxically low ANP levels. These findings may indicate that a relative deficiency of ANP (an inhibitor of AVP secretion) in the setting of chronically increased LA pressure may contribute to AVP excess.

**Keywords** Arginine vasopressin; Heart failure with preserved ejection fraction; Left ventricular hypertrophy; Left atrium; Atrial natriuretic peptide

Received: 29 January 2018; Accepted: 30 May 2018

\*Correspondence to: Julio A. Chirinos, MD, PhD, Perelman Center for Advanced Medicine, South Tower, Rm. 11-138, 3400 Civic Center Blvd., Philadelphia, PA 19104, USA. Tel: 215 573 6606; Fax: 215 746 7415. Email: julio.chirinos@uphs.upenn.edu

†Julio A. Chirinos and Mayank Sardana have equal contribution in this study.

## Introduction

The arginine vasopressin (AVP) pathway is important in the regulation of sodium and water metabolism, as well as vascular homeostasis.<sup>1</sup> Arginine vasopressin exerts a wide variety of effects on the heart, vascular smooth muscle, vascular endothelium, platelets, and kidneys. V1a receptors

mediate vasoconstriction, platelet aggregation, myocardial hypertrophy, and fibrosis, whereas activation of V2 receptors in renal collecting ducts mediates the antidiuretic effects of AVP.<sup>2</sup>

Excessive activation of the AVP pathway has been considered a maladaptive response to heart failure (HF), contributing to volume retention.<sup>1</sup> Circulating levels of AVP have been

studied in HF with reduced ejection fraction (HFrEF)<sup>3</sup> and shown to correlate with the severity of left ventricular (LV) dysfunction<sup>4</sup> and with clinical outcomes.<sup>5</sup> However, little data are available regarding the role of AVP in HF with preserved ejection fraction (HFpEF). Recent studies demonstrated increased circulating copeptin levels (a surrogate for AVP release) in subjects with HFpEF.<sup>6,7</sup> However, the correlates of increased AVP levels in HFpEF are not well understood. In particular, whether increased AVP levels are associated with LV remodelling in HFpEF and HFrEF is unknown. Similarly, little is known about the association between AVP levels and atrial natriuretic peptide (ANP) levels, which are released in response to increased atrial stretch and inhibit AVP release.<sup>8</sup>

In this study, we aimed to (i) compare levels of plasma AVP between subjects with HFpEF and HFrEF and subjects without HF; (ii) assess the association between AVP, ANP levels, and left atrial (LA) volume; (iii) assess the association between AVP and LV remodelling; and (iv) assess the association between AVP and the risk of incident cardiovascular death or HF admission.

## Methods

We prospectively enrolled a convenience sample of patients referred for a cardiac magnetic resonance imaging (MRI) study at the Corporal Michael J. Crescenz VA Medical Center. The protocol was approved by the Philadelphia VA Medical Center Institutional Review Board, and all subjects provided written informed consent.

Heart failure with reduced ejection fraction was defined as a symptomatic HF in the presence of an LV ejection fraction (LVEF) < 50%. Heart failure with preserved ejection fraction was defined as (i) New York Heart Association Class II–IV symptoms consistent with HF, in the absence of significant aortic or mitral stenosis; (ii) LVEF > 50%; and (iii) a mitral E wave to annular  $e'$  ratio > 14,<sup>9</sup> or at least two 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) LA volume index > 34 mL/m<sup>2</sup> of body surface area; (d) N-terminal pro B-type natriuretic peptide level > 200 pg/mL; and (e) LV mass index > 149 g/m<sup>2</sup> in men and 122 g/m<sup>2</sup> in women (measured by cardiac MRI). Subjects without HF had an LVEF > 50%, no significant valvular disease, and no symptoms and signs consistent with HF.

Key exclusion criteria were as follows: (i) claustrophobia; (ii) presence of metallic objects or implanted medical devices in body; (iii) conditions that could make the interpretation of MRI less accurate and/or unreliable (i.e. arrhythmia such as atrial fibrillation affecting cardiac gating and inability to hold breath for the cardiac MRI acquisitions); and (iv) known

infiltrative or hypertrophic cardiomyopathy, or extra-cardiac amyloidosis or sarcoidosis.

## Measurement of serum arginine vasopressin and atrial natriuretic peptide

Venous plasma samples were obtained at the time of enrolment and stored at  $-80^{\circ}\text{C}$  for batch analysis. Arginine vasopressin and ANP were measured with enzyme-linked immunosorbent assay technique (Cayman Chemical, Ann Arbor, MI, USA).

## Measurement of central blood pressure

We measured central blood pressure via carotid arterial tonometry performed in the supine position immediately after the MRI, using a high-fidelity Millar applanation tonometer (SPT-301; Millar Instruments, Houston, TX, USA) and a dedicated acquisition platform (Sphygmocor device; Atcor Medical, Sydney, Australia). This approach does not require the use of a generalized transfer function, because the carotid pressure waveform is a direct surrogate of aortic pressure.

## Cardiac magnetic resonance imaging

We measured LV mass and volume and LA volume, using a 1.5 Tesla whole-body MRI scanner (Avanto or Espree; Siemens, Malvern, PA, USA) equipped with a phase-array cardiac coil.

Left ventricular volumes (end-diastolic and end-systolic volumes) and function (ejection fraction) were measured using steady-state free-precession cine imaging. Typical acquisition parameters were repetition time (TR) = 30.6 ms; echo time (TE) = 1.3 ms; slice thickness = 8 mm; phases = 30; parallel image (IPAT) factor = 2; and matrix size = 192 × 192. CMR42 software (Circle CVI, Calgary, AB, Canada) was utilized to manually trace the LV short-axis cine images at end of diastole and systole. Left ventricular mass was calculated as the difference between epicardial and endocardial volumes, multiplied by the myocardial density. Left ventricular mass was normalized for body height raised to the allometric power of 1.7.<sup>10</sup> Left atrial volume was calculated by averaging the volumes measured end-systole by manually tracing the LA endocardial border in the apical two-chamber and four-chamber views.

## Statistical methods

Continuous and categorical variables were compared between the groups using analysis of variance and chi-squared

tests, respectively. Multivariable linear regression models were utilized to assess HFpEF and HFrEF as predictors of AVP levels, with and without adjusting for potential confounders. When required, Box–Cox transformation was applied to normalize regression model residuals. A second set of multivariable linear regression models were utilized to assess the association between ANP and AVP. Multivariable linear regression was also utilized to assess whether AVP was associated with LV mass index. We present standardized regression coefficients for easier comparison of the magnitude of the effect of various predictors on the dependent variable in regression models. The association between AVP levels and the risk of a composite endpoint of incident hospitalized HF or cardiovascular death was assessed with proportional hazards (Cox) regression. All tests were two-tailed. Statistical significance was defined as a  $P$ -value  $\leq 0.05$ . We used SPSS v24 for Mac (IBM, Chicago, IL, USA) and Matlab v2016b (The Mathworks; Natick, MA, USA) to perform statistical analyses.

## Results

Baseline characteristics of our participants with HFpEF and HFrEF and no HF are presented and compared in *Table 1*.

### Association of arginine vasopressin and heart failure with preserved ejection fraction

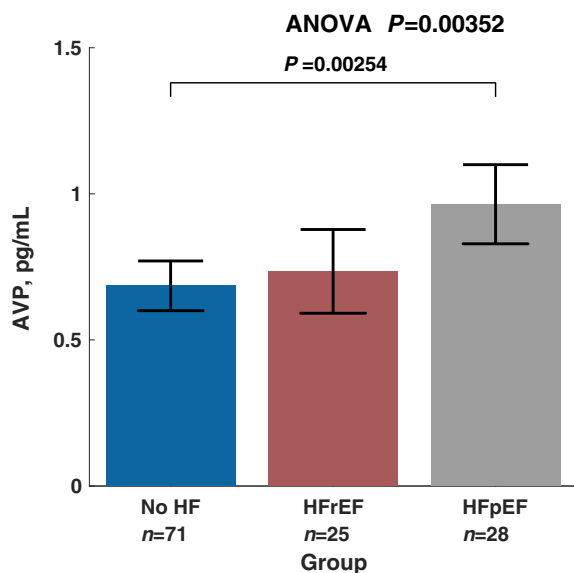
*Figure 1* shows mean AVP levels in patients without HF, HFpEF, and HFrEF. There were significant between-group differences in AVP levels (analysis of variance  $P = 0.0002$ ). Arginine vasopressin levels were significantly greater in HFpEF [0.96 pg/mL; 95% confidence interval (CI) = 0.83–1.1 pg/mL] compared with controls (0.69 pg/mL; 95% CI = 0.6–0.77 pg/mL;  $P$  for pairwise comparison = 0.0003). In *post hoc* pairwise comparisons, AVP levels among subjects with HFrEF (0.73 pg/mL; 95%

**Table 1** General characteristics of study participants

	No HF (n = 71)	HFrEF (n = 25)	HFpEF (n = 28)	P-value
Age	61.55 (58.97 to 64.13)	65.2 (60.85 to 69.55)	63.61 (59.49 to 67.72)	0.34
Male sex	64 (90.14%)	24 (96.00%)	24 (85.71%)	0.45
Race/ethnicity				
White	34 (47.89%)	14 (56.00%)	8 (28.57%)	0.10
African American	33 (46.48%)	11 (44.00%)	20 (71.43%)	0.057
Other	4 (5.63%)	0 (0.00%)	0 (0.00%)	0.21
Body mass index (kg/m <sup>2</sup> )	30.3 (28.84 to 31.77)	29.53 (27.07 to 32)	35.7 (33.36 to 38.03)	0.0003
Hypertension	59 (83.10%)	21 (84.00%)	27 (96.43%)	0.21
Coronary artery disease	19 (26.76%)	21 (84.00%)	10 (35.71%)	<0.0001
Diabetes mellitus	40 (56.34%)	14 (56.00%)	22 (78.57%)	0.11
History of atrial fibrillation	2 (2.82%)	1 (4.00%)	1 (3.57%)	0.95
History of CVA or TIA	10 (14.08%)	7 (28.00%)	4 (14.29%)	0.26
Peripheral vascular disease	6 (8.45%)	4 (16.00%)	7 (25.00%)	0.09
Current smoker	15 (21.13%)	8 (32.00%)	5 (17.86%)	0.42
Beta-blocker use	29 (40.85%)	22 (88.00%)	22 (78.57%)	<0.0001
ACE inhibitor use	36 (50.70%)	18 (72.00%)	17 (60.71%)	0.17
ARB use	5 (7.04%)	2 (8.00%)	6 (21.43%)	0.10
Furosemide use	2 (2.82%)	14 (56.00%)	16 (57.14%)	<0.0001
Spironolactone use	1 (1.41%)	2 (8.00%)	1 (3.57%)	0.27
Calcium channel blocker use	25 (35.21%)	6 (24.00%)	11 (39.29%)	0.47
Thiazide use	19 (26.76%)	7 (28.00%)	10 (35.71%)	0.67
eGFR (mL/m <sup>2</sup> )	83.88 (78.04 to 89.73)	79.76 (69.91 to 89.61)	79.32 (70.02 to 88.63)	0.63
Systolic BP (mmHg)	144.5 (140 to 149.1)	141 (133.3 to 148.7)	154.5 (147.2 to 161.8)	0.029
Diastolic BP (mmHg)	83.61 (81 to 86.3)	80.2 (75.8 to 84.7)	87.5 (83.3 to 91.7)	0.069
Central systolic BP (mmHg)	136.7 (131.29 to 142.1)	136.6 (127.7 to 145.6)	150.8 (141.8 to 159.8)	0.026
NYHA Class III–IV	0	1 (4.17%)	1 (3.57%)	0.25
LV EDV (mL)	145 (132.8 to 157.2)	231.9 (211.4 to 252.5)	170.3 (150.8 to 189.7)	<0.0001
LV mass (g)	145.8 (135.2 to 156.4)	186.6 (168.7 to 204.5)	182 (165.0 to 198.9)	<0.0001
LVEF (%)	60.07 (57.79 to 62.34)	35.48 (31.65 to 39.32)	63.31 (59.69 to 66.93)	<0.0001
LV mass index (g/m <sup>1.7</sup> )	56.29 (52.23 to 60.36)	70.82 (63.96 to 77.68)	71.46 (64.98 to 77.94)	<0.0001
LVEDV index (mL/m <sup>2</sup> )	48.81 (44.8 to 52.83)	77.14 (70.37 to 83.91)	57.81 (51.42 to 64.21)	<0.0001
LA volume (mL)	59.9 (52.8 to 67)	79.1 (61.9 to 96.3)	77 (62.3 to 91.6)	0.013
LA volume index (mL/m <sup>2</sup> )	28.3 (25 to 31.7)	36.8 (28.9 to 44.6)	33.9 (27.6 to 40.3)	0.041
Mitral inflow A wave velocity (cm/s)	74.1 (68.6 to 79.5)	61.9 (52.8 to 71)	72.3 (64.5 to 80.1)	0.08
Mitral inflow E wave velocity (cm/s)	69.3 (63.1 to 75.6)	65.6 (55.1 to 76)	82.9 (74.2 to 91.7)	0.02
Mitral inflow E wave deceleration time (ms)	214 (195 to 233)	214 (181 to 247)	210 (182 to 238)	0.98
E/e'	9.6 (8.3 to 10.8)	11.7 (9.1 to 14.3)	12.4 (10.1 to 14.7)	0.04

ACE, angiotensin convertase enzyme; ARB, angiotensin receptor blocker; BP, blood pressure; CVA, cerebrovascular accident; E/e', mitral inflow to mitral annular tissue velocity ratio; eGFR, estimated glomerular filtration rate; HF, heart failure; HFpEF, heart failure with preserved ejection fraction; HFrEF, heart failure with reduced ejection fraction; LA, left atrial; LV, left ventricular; LVEDV, left ventricular end-diastolic volume; LVEF, left ventricular ejection fraction; NYHA, New York Heart Association; TIA, transient ischaemic attack.

**Figure 1** Comparison of arginine vasopressin (AVP) levels between subjects without heart failure (HF), subjects with heart failure with reduced ejection fraction (HFrEF), and heart failure with preserved ejection fraction (HFpEF).



CI = 0.59–0.88 pg/mL) did not significantly differ from any of the other two groups.

We also assessed the presence of HFpEF or HFrEF as predictors of AVP levels in unadjusted and adjusted linear regression models. In unadjusted analyses, the presence of HFpEF (standardized  $\beta = 0.31$ ; 95% CI = 0.13–0.48;  $P = 0.0001$ ) was associated with higher AVP levels. In a multivariable linear regression model adjusted for age, sex, race, systolic blood pressure, diabetes mellitus, the use of angiotensin convertase enzyme inhibitors, angiotensin receptor blockers, furosemide, spironolactone, thiazide diuretics, calcium channel blockers, beta-blockers, and estimated glomerular filtration rate, the presence of HFpEF was independently associated with higher AVP levels (standardized  $\beta = 0.27$ ; 95% CI = 0.04–0.5;  $P = 0.025$ ).

### Atrial natriuretic peptides and left atrial volume: association with arginine vasopressin

Plasma ANP was negatively associated with AVP (standardized  $\beta = 0.25$ ; 95% CI =  $-0.43$  to  $-0.065$ ;  $P = 0.008$ ). *Figure 2* (bottom left panel) shows mean ANP levels among subjects above and below the median value of AVP (0.64 pg/mL). Atrial natriuretic peptide levels were 828 (95% CI = 706–950) and 588 (95% CI = 501–674) pg/mL in subjects below and above the median value of AVP, respectively ( $P = 0.008$ ). In a linear regression model that included ANP and LA volume, LA volume was positively associated (standardized  $\beta = 0.21$ ; 95% CI = 0.03–0.40;  $P = 0.022$ ), whereas ANP was negatively

associated (standardized  $\beta = -0.19$ ; 95% CI =  $-0.37$  to  $-0.01$ ;  $P = 0.04$ ) with AVP levels (*Figure 2*, top panel). The ANP/LA volume ratio was higher (12.3; 95% CI = 9.6–15.1) in subjects below vs. those above (8.5; 95% CI = 6.6–10.4) the median value of AVP ( $P = 0.011$ ; *Figure 2*; bottom right panel).

In a multivariable model that adjusted for HF group membership, race, sex, age, systolic blood pressure, diabetes mellitus, the use of angiotensin convertase enzyme inhibitors, angiotensin receptor blockers, furosemide, estimated glomerular filtration rate, and New York Heart Association Class III–IV (*Table 2*), the two independent correlates of a higher AVP were the presence of HFpEF (standardized  $\beta = 0.32$ ; 95% CI = 0.09–0.56;  $P = 0.0073$ ) and the ANP/LA volume ratio (standardized  $\beta = -0.23$ ; 95% CI =  $-0.42$  to  $-0.04$ ;  $P = 0.0196$ ).

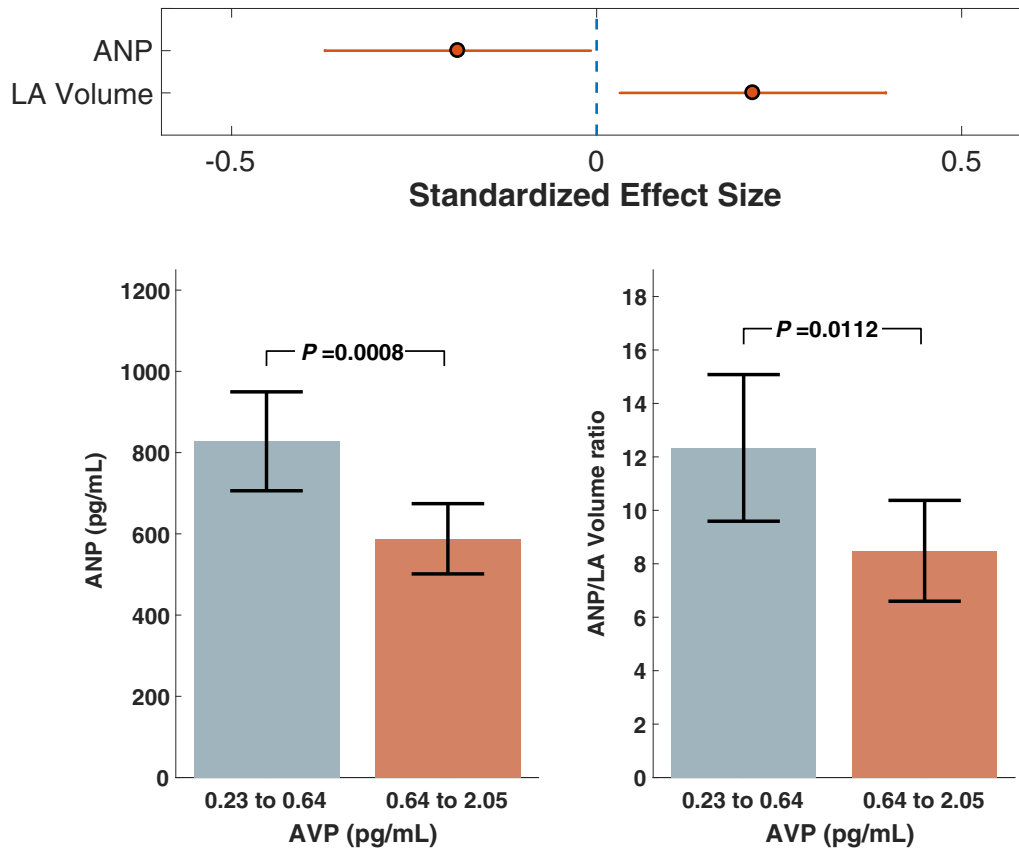
### Association of arginine vasopressin levels and left ventricular hypertrophy

In unadjusted analyses, AVP levels were directly associated with LV mass (standardized  $\beta = 0.29$ ; 95% CI = 0.12–0.46;  $P = 0.001$ , *Figure 3*, upper panel). In a model that adjusted for HF status, age, sex, race, glomerular filtration rate and central systolic blood pressure (*Figure 3*, lower panel), AVP was a significant independent predictor of LV mass (standardized  $\beta = 0.24$ ; 95% CI = 0.07–0.40;  $P = 0.005$ ). Similarly, after further adjustment for the use of angiotensin convertase enzyme inhibitors, angiotensin receptor blockers, furosemide, spironolactone, thiazide diuretics, calcium channel blockers, and beta-blockers, AVP continued to be directly associated with greater LV mass index (standardized  $\beta = 0.26$ ; 95% CI = 0.09–0.43;  $P = 0.003$ ).

### Arginine vasopressin as a predictor of outcomes

During a median follow-up of 1507 days (~4.12 years), 15 subjects had an HF admission, and 10 died of cardiovascular causes. *Figure 4* shows the Kaplan–Meier event-free survival curves for subjects stratified by the median value of AVP. Survival was significantly lower in subjects with higher AVP. In a proportional hazards regression model, AVP was a significant predictor of an increased risk of incident cardiovascular death or HF admission [standardized hazard ratio (HR) = 1.61; 95% CI = 1.13–2.29;  $P = 0.008$ ]. Similarly, AVP predicted the endpoint after adjustment for age, gender, and HF group membership (standardized HR = 1.65; 95% CI = 1.10–2.49;  $P = 0.016$ ). The association between AVP and incident cardiovascular death or hospitalized HF was also independent of LA volume index, LV mass, and ANP (standardized HR = 1.82; 95% CI = 1.12–2.96;  $P = 0.015$ ). In an analysis that included only subjects with HF (HFpEF or HFrEF), AVP predicted

**Figure 2** Association between atrial natriuretic peptide (ANP) and left atrial (LA) volume vs. arginine vasopressin (AVP). High AVP was associated with lower ANP levels. The bottom left panel shows mean ANP levels in subjects above vs. below the median value of AVP. In a linear regression model, a low ANP and a higher LA volume were independent predictors of AVP (top panel). Subjects with AVP levels above the median value demonstrated lower ANP/LA volume ratios (right bottom panel).



**Table 2** Correlates of AVP in multivariable linear regression

	Standardized estimate	95% CI, lower bound	95% CI, upper bound	<i>P</i> -value
ANP/LA volume ratio	-0.23	-0.42	-0.04	0.0196
HFpEF	0.32	0.09	0.56	0.0073
HFrEF	0.01	-0.21	0.23	0.8985
Age	-0.10	-0.31	0.12	0.3670
Male sex	0.16	-0.03	0.35	0.1022
African American ethnicity	0.16	-0.03	0.35	0.1036
Systolic blood pressure	-0.06	-0.28	0.15	0.5567
Diabetes mellitus	-0.12	-0.33	0.08	0.2268
ACE inhibitor use	0.06	-0.14	0.26	0.5292
ARB use	-0.05	-0.26	0.16	0.6208
Furosemide use	0.16	-0.08	0.40	0.1868
eGFR	-0.05	-0.25	0.15	0.6338
NYHA Class III-IV	-0.12	-0.31	0.07	0.22421

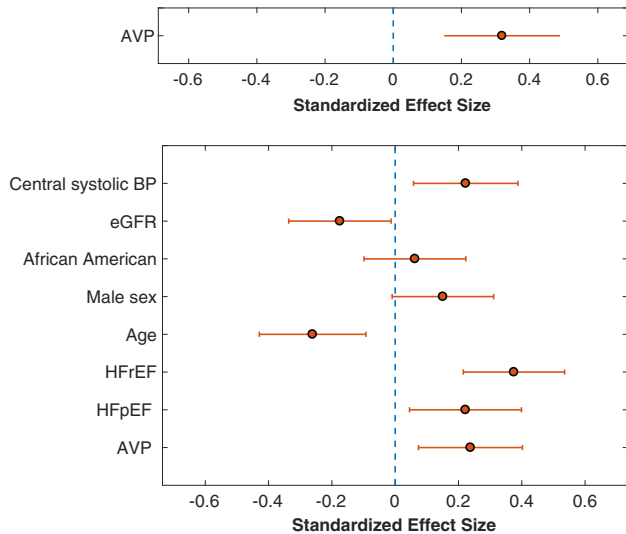
ACE, angiotensin convertase enzyme; ANP, atrial natriuretic peptide; ARB, angiotensin receptor blocker; AVP, arginine vasopressin; CI, confidence interval; eGFR, estimated glomerular filtration rate; HFpEF, heart failure with preserved ejection fraction; HFrEF, heart failure with reduced ejection fraction; LA, left atrial; NYHA, New York Heart Association.

incident cardiovascular death or HF admissions (standardized HR = 1.72; 95% CI = 1.09–2.72; *P* = 0.021).

## Discussion

In this study, we report that HFpEF is associated with increased plasma AVP, even after adjustment for potential confounders. We report, for the first time, that AVP levels are independently associated with the combination of greater LA volume and paradoxically low ANP levels. This finding is consistent with the physiological regulation of AVP by ANP demonstrated in animal models and raise the hypothesis that atrial natriuretic peptide deficiency in the setting of increased LA pressure is related to AVP excess, which should be tested in future studies. We demonstrate that AVP independently correlates with LV hypertrophy measured by MRI. Finally, we show that greater AVP levels are associated with an increased risk of adverse outcomes, independent of ANP, LA volume, and LV mass. Our findings contribute to

**Figure 3** Predictors of left ventricular (LV) mass by linear regression. The top panel shows an unadjusted model showing the association between arginine vasopressin (AVP) and LV mass. The bottom panel shows a multivariable model demonstrating independent correlates of LV mass. BP, blood pressure; eGFR, estimated glomerular filtration rate; HFpEF, heart failure with preserved ejection fraction; HFrEF, heart failure with reduced ejection fraction.



our understanding of the correlates of circulating AVP excess and its clinical relevance in human HF.

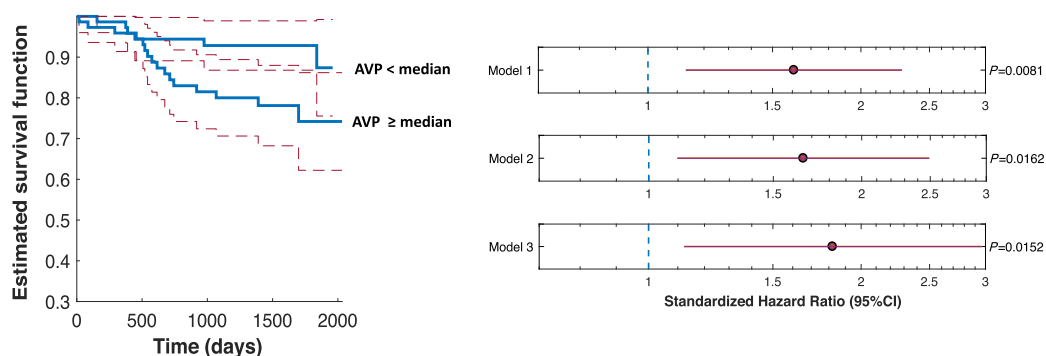
### Arginine vasopressin in heart failure

Arginine vasopressin is a nonapeptide synthesized by the hypothalamus, and it is released into the circulation from the neurohypophysis in response to various osmotic and non-osmotic stimuli (intravascular hypovolaemia).<sup>2</sup> Initial studies in patients with HFrEF and asymptomatic LV dysfunction reported that AVP levels are inappropriately elevated in these

conditions (in response to non-osmotic stimuli).<sup>3,4,11</sup> Secretion of AVP can lead to deleterious effects in HF through its vasoconstrictive (V1a receptors) and free water retaining properties (V2 receptors), as well as through its hypertrophy and fibrosis promoting effects on the myocardium (V1a receptors) leading to adverse cardiac remodelling.<sup>1</sup> Whereas great attention has been placed on V2-related effects on hyponatraemia in advanced HF, much less is known about the role of vascular and myocardial V1a effects. Finally, little is known about the relationship between ANP and AVP levels, despite its well-known physiological cross-regulation.<sup>8</sup>

To the best of our knowledge, no prior study has reported the comparative association of plasma AVP with HFpEF vs. HFrEF. We observed that in adjusted models, mean levels of AVP were significantly higher in HFpEF as compared with HFrEF and the control group. Of note, average AVP levels in our participants with HFrEF were lower than those previously reported in the older studies,<sup>3,4</sup> However, more recent studies have reported similar results as ours.<sup>5</sup> In the Efficacy of Vasopressin Antagonism in Heart Failure Outcome Study with Tolvaptan (EVEREST), 59% of the participants had AVP levels below the lower detection limit of the assay, and another 19% had levels within normal range.<sup>5</sup> This difference might be related to (i) increased utilization of guideline directed medical therapy in recent studies<sup>12</sup> and (ii) differences in assay sensitivity.<sup>13</sup> Copeptin is the C-terminal peptide of the precursor protein to AVP and is secreted in equimolar amounts as AVP in the circulation.<sup>14</sup> Mainly due to the short half-life of AVP, copeptin has been utilized as a surrogate marker of AVP activity in recent investigations.<sup>6,7</sup> Data from these investigations suggest that higher copeptin levels are associated with an increased risk of adverse outcomes in both unselected HF<sup>6,7</sup> and selected samples with HFpEF.<sup>15</sup> Contrary to our findings (higher AVP in HFpEF compared with HFrEF), Loncar *et al.*<sup>16</sup> reported that copeptin levels were significantly higher in HFrEF compared with HFpEF. However, their study enrolled participants with recent HF exacerbation, which might have led to a higher degree of

**Figure 4** Left: Kaplan–Meier survival curves for cardiovascular death or heart failure hospitalization among subjects stratified according to the median value of arginine vasopressin (AVP). Dashed red lines represent 95% confidence interval (CI).



neurohormonal activation (and AVP levels) when compared with the participants with chronic HF enrolled in our study.

### Arginine vasopressin excess and left ventricular mass

Prior studies in animal models have demonstrated that the activation of V1a receptors by AVP leads to structural changes in cardiac myocytes and fibroblasts leading to an increase in myocardial mass (LV hypertrophy).<sup>17,18</sup> Tozawa *et al.*<sup>19</sup> reported that in spontaneously hypertensive rats treated with an angiotensin convertase enzyme inhibitor (delapril), there was a positive direct association between LV mass and AVP levels. In a cross-sectional analysis of 706 middle-aged participants without cardiovascular comorbidities, Bhandari *et al.*<sup>20</sup> reported a weak direct correlation of copeptin levels with indexed LV mass. In a prospective investigation of middle-aged participants, Strand *et al.*<sup>21</sup> demonstrated that higher baseline AVP levels predicted the development of LV hypertrophy at 20 years of follow-up in 17 participants who were diagnosed with hypertension in interim. We hereby report a direct independent association between AVP levels and LV mass determined with cardiac MRI, independent of the presence or absence of HF and multiple potential confounders, including renal function and central systolic blood pressure. Taken together, available animal studies and human data support a role for AVP in the development of LV hypertrophy, an important cardiovascular phenotype involved in the pathogenesis of HF.<sup>22</sup>

### Arginine vasopressin, atrial natriuretic peptide, and left atrial volume

Despite the importance of AVP and its role in hyponatraemia in advanced HF, little is known about the non-osmotic triggers for its release in humans with and without HF. A role for baroreceptor desensitization has been proposed as a cause for dysregulated AVP release.<sup>23</sup> However, human studies assessing the association between the ANP axis and AVP levels are scarce. Atrial natriuretic peptide, a member of the natriuretic peptide family, is released in response to atrial stretch and exerts a variety of biological effects such as natriuresis, vasodilation, and inhibition of renin–angiotensin–aldosterone pathway.<sup>24</sup> Animal models have also established an inhibitory role for ANP on AVP release.<sup>25</sup> Vascular and intracerebral administration of ANP inhibits the release of AVP,<sup>8</sup> and ANP binding sites after systemic administration of ANP have been identified in the posterior pituitary.<sup>25</sup> Interestingly, in our study, LA enlargement with paradoxically low ANP levels was associated with increased AVP levels. Although physiological considerations from animal models suggest that a deficient ANP axis may causally contribute to

such excess, further studies are required to establish this mechanism in human HF and to determine whether strategies aimed at enhancing natriuretic peptide activity blunt AVP excess and its deleterious consequences. Our findings are consistent with the recently proposed paradigm that a deficiency of ANP and/or increased target organ resistance to ANP actions contribute to LV remodelling<sup>26</sup> and HF.<sup>27</sup>

Our study also demonstrates that increased AVP levels are associated with an increased risk of incident cardiovascular death or hospitalized HF. This association was highly significant and was independent of ANP levels, LV mass, and LV enlargement measured by cardiac MRI, supporting the clinical importance of AVP.

Our study should be interpreted in context of its strengths and limitations. We utilized cardiac MRI, which provides highly accurate measurements of chamber volumes and function. However, we acknowledge that our study has limitations. Our findings are observational in nature, and this cannot prove causal associations. Although most analyses were based on well-established quantitative traits, our sample size and the number of prospective events during follow-up was relatively small, and thus, only limited adjustments were possible to avoid overfitting of proportional hazards models; the low number of events also precluded stratified analyses by HF subtype. Residual confounding could still be present, and larger future studies should study this further. Lastly, we utilized convenience sampling of subjects referred for a cardiac MRI study at a Veteran Affairs Medical Center. Thus, the majority of the participants were male and may not be representative of AVP levels seen in unselected HF populations. We also applied various exclusion criteria. An important exclusion criterion was atrial fibrillation (because atrial fibrillation complicates measurements of LV mass by segmented cine MRI). Therefore, extrapolation of our findings to patients with advanced atrial disease and/or atrial arrhythmia cannot be made. We did not assess the potential role of the sympathetic nervous system, which is physiologically linked to AVP secretion and subject to inhibition by ANP. The interactions between AVP, ANP, and the sympathetic nervous system in HF (particularly HFpEF) should be the focus of future research. Our population exhibited relatively favourable outcomes, and our results may not extrapolate to patients with more advanced disease.

## Conclusions

Increased plasma AVP is seen in subjects with HFpEF, even after adjustment for potential confounders. Arginine vasopressin levels are independently associated with a greater LA volume and also paradoxically, with lower ANP levels. These findings are consistent with the known inhibitory effect of

ANP on AVP secretion. High plasma AVP was independently associated with LV mass measured by cardiac MRI. High AVP was predictive of an increased risk of cardiovascular death and HF admissions, independently of LV mass, LA volume, and ANP levels. Our findings demonstrate the clinical importance of AVP in HFpEF and suggest that a relative deficiency of ANP in the setting of increased LA pressure is associated with AVP excess. Further studies are required to establish whether this association is causal and to assess whether strategies aimed at potentiating the ANP axis may ameliorate AVP excess and its deleterious effects.

## Conflict of interest

J.A.C. has received consulting honoraria from Bristol-Myers Squibb, OPKO Healthcare, Fukuda Denshi, Microsoft, Sanifit, Pfizer, Ironwood Pharmaceuticals, Vital Labs, and Merck. He

received research grants from the National Institutes of Health, American College of Radiology Network, Fukuda Denshi, Bristol-Myers Squibb, Microsoft, and CVRx Inc. and device loans from AtCor Medical. J.A.C. is named as inventor in a University of Pennsylvania patent application for the use of inorganic nitrates/nitrites for the treatment of heart failure and preserved ejection fraction.

## Funding

This study was supported by the National Institutes of Health grants R56HL-124073-01A1 (J.A.C.), R01 HL 121510-01A1 (J.A.C.), and 5-R21-AG-043802-02 (J.A.C.) and a VISN-4 research grant from the Department of Veterans Affairs (J.A.C.) and in part by the Intramural Research Program, National Institute on Aging, National Institutes of Health.

## References

- Chatterjee K. Neurohormonal activation in congestive heart failure and the role of vasopressin. *Am J Cardiol* 2005; **95**: 8B–13B.
- Holmes CL, Landry DW, Granton JT. Science review: vasopressin and the cardiovascular system part 2—clinical physiology. *Crit Care* 2004; **8**: 15–23.
- Finley JJ, Konstam MA, Udelson JE. Arginine vasopressin antagonists for the treatment of heart failure and hyponatremia. *Circulation* 2008; **118**: 410–421.
- Francis GS, Benedict C, Johnstone DE, Kirlin PC, Nicklas J, Liang CS, Kubo SH, Rudin-Toretzky E, Yustuf S. Comparison of neuroendocrine activation in patients with left ventricular dysfunction with and without congestive heart failure. A substudy of the Studies of Left Ventricular Dysfunction (SOLVD). *Circulation* 1990; **82**: 1724–1729.
- Lanfang DE, Sabbah HN, Goldsmith SR, Greene SJ, Ambrosy AP, Fought AJ, Kwasny MJ, Swedberg K, Yancy CW, Konstam MA, Maggioni AP, Zannad F, Gheorghide M, Investigators Et. Association of arginine vasopressin levels with outcomes and the effect of V2 blockade in patients hospitalized for heart failure with reduced ejection fraction: insights from the EVEREST trial. *Circ Heart Fail* 2013; **6**: 47–52.
- Alehagen U, Dahlstrom U, Rehfeld JF, Goetze JP. Association of copeptin and N-terminal proBNP concentrations with risk of cardiovascular death in older patients with symptoms of heart failure. *JAMA* 2011; **305**: 2088–2095.
- Neuhold S, Huelsmann M, Strunk G, Stoiser B, Struck J, Morgenthaler NG, Bergmann A, Moertl D, Berger R, Pacher R. Comparison of copeptin, B-type natriuretic peptide, and amino-terminal pro-B-type natriuretic peptide in patients with chronic heart failure: prediction of death at different stages of the disease. *J Am Coll Cardiol* 2008; **52**: 266–272.
- Matsukawa T, Miyamoto T. Angiotensin II-stimulated secretion of arginine vasopressin is inhibited by atrial natriuretic peptide in humans. *Am J Physiol Regul Integr Comp Physiol* 2011; **300**: R624–R629.
- Nagueh SF, Smiseth OA, Appleton CP, Byrd BF 3rd, Dokainish H, Edvardsen T, Flachskampf FA, Gillebert TC, Klein AL, Lancellotti P, Marino P, Oh JK, Popescu BA, Waggoner AD. Recommendations for the evaluation of left ventricular diastolic function by echocardiography: an update from the American Society of Echocardiography and the European Association of Cardiovascular Imaging. *J Am Soc Echocardiogr* 2016; **29**: 277–314.
- Chirinos JA, Segers P, De Buyzere ML, Kronmal RA, Raja MW, De Bacquer D, Claessens T, Gillebert TC, St John-Sutton M, Rietzschel ER. Left ventricular mass: allometric scaling, normative values, effect of obesity, and prognostic performance. *Hypertension* 2010; **56**: 91–98.
- Udelson JE, McGrew FA, Flores E, Ibrahim H, Katz S, Koshkarian G, O'Brien T, Kronenberg MW, Zimmer C, Orlandi C, Konstam MA. Multicenter, randomized, double-blind, placebo-controlled study on the effect of oral tolvaptan on left ventricular dilation and function in patients with heart failure and systolic dysfunction. *J Am Coll Cardiol* 2007; **49**: 2151–2159.
- Yancy CW, Jessup M, Bozkurt B, Butler J, Casey DE Jr, Drazner MH, Fonarow GC, Geraci SA, Horwich T, Januzzi JL, Johnson MR, Kasper EK, Levy WC, Masoudi FA, McBride PE, McMurray JJ, Mitchell JE, Peterson PN, Riegel B, Sam F, Stevenson LW, Tang WH, Tsai EJ, Wilkoff BL. 2013 ACCF/AHA guideline for the management of heart failure: executive summary: a report of the American College of Cardiology Foundation/American Heart Association Task Force on practice guidelines. *Circulation* 2013; **128**: 1810–1852.
- Leng G, Sabatier N. Measuring oxytocin and vasopressin: bioassays, immunoassays and random numbers. *J Neuroendocrinol* 2016; **28**.
- Morgenthaler NG. Copeptin: a biomarker of cardiovascular and renal function. *Congest Heart Fail* 2010; **16**: S37–S44.
- Hage C, Lund LH, Donal E, Daubert JC, Linde C, Mellbin L. Copeptin in patients with heart failure and preserved ejection fraction: a report from the prospective KaRen-study. *Open Heart* 2015; **2**: e000260.
- Loncar G, von Haehling S, Tahirovic E, Inkrot S, Mende M, Sekularac N, Lainscak M, Apostolovic S, Putnikovic B, Edelmann F, Wachter R, Dimkovic S, Waagstein F, Gelbrich G, Dungen HD. Effect of beta blockade on natriuretic



- peptides and copeptin in elderly patients with heart failure and preserved or reduced ejection fraction: results from the CIBIS-ELD trial. *Clin Biochem* 2012; **45**: 117–122.
17. Fukuzawa J, Haneda T, Kikuchi K. Arginine vasopressin increases the rate of protein synthesis in isolated perfused adult rat heart via the V1 receptor. *Mol Cell Biochem* 1999; **195**: 93–98.
  18. Fan YH, Zhao LY, Zheng QS, Dong H, Wang HC, Yang XD. Arginine vasopressin increases iNOS–NO system activity in cardiac fibroblasts through NF- $\kappa$ B activation and its relation with myocardial fibrosis. *Life Sci* 2007; **81**: 327–335.
  19. Tozawa M, Takishita S, Muratani H, Fukiyama K. Non-corresponding effects of an angiotensin-converting enzyme inhibitor on cardiac and vascular hypertrophy in spontaneously hypertensive rats. *Hypertens Res* 2000; **23**: 483–490.
  20. Bhandari SS, Loke I, Davies JE, Squire IB, Struck J, Ng LL. Gender and renal function influence plasma levels of copeptin in healthy individuals. *Clin Sci (Lond)* 2009; **116**: 257–263.
  21. Strand AH, Gudmundsdottir H, Fossum E, Os I, Bjornerheim R, Kjeldsen SE. Arterial plasma vasopressin and aldosterone predict left ventricular mass in men who develop hypertension over 20 years. *J Clin Hypertens (Greenwich)* 2007; **9**: 365–371.
  22. de Simone G, Gottdiener JS, Chinali M, Maurer MS. Left ventricular mass predicts heart failure not related to previous myocardial infarction: the Cardiovascular Health Study. *Eur Heart J* 2008; **29**: 741–747.
  23. Trimarco B, De Luca N, De Simone A, Volpe M, Ricciardelli B, Lembo G, Condorelli M. Impaired control of vasopressin release in hypertensive subjects with cardiac hypertrophy. *Hypertension* 1987; **10**: 595–602.
  24. Kasama S, Furuya M, Toyama T, Ichikawa S, Kurabayashi M. Effect of atrial natriuretic peptide on left ventricular remodelling in patients with acute myocardial infarction. *Eur Heart J* 2008; **29**: 1485–1494.
  25. Gerstberger R, Schutz H, Luther-Dyroff D, Keil R, Simon E. Inhibition of vasopressin and aldosterone release by atrial natriuretic peptide in conscious rabbits. *Exp Physiol* 1992; **77**: 587–600.
  26. Ibebuogu UN, Gladysheva IP, Houg AK, Reed GL. Decompensated heart failure is associated with reduced corin levels and decreased cleavage of pro-atrial natriuretic peptide. *Circ Heart Fail* 2011; **4**: 114–120.
  27. Diez J. Chronic heart failure as a state of reduced effectiveness of the natriuretic peptide system: implications for therapy. *Eur J Heart Fail* 2017; **19**: 167–176.