Biomarkers in persistent AF and heart failure: impact of catheter ablation compared with rate-control

Short title: Biomarkers after AF ablation in heart failure

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Abstract

Background

To investigate the effects of catheter-ablation and rate-control strategies on cardiac and inflammatory biomarkers in patients with heart failure and persistent atrial fibrillation (AF).

Methods

Patients were recruited from the ARC-HF trial (NCT00878384), which compared ablation with rate-control for persistent AF in heart failure. B-type natriuretic peptide (BNP), mid-regional pro-atrial natriuretic peptide(MR-proANP), Apelin and Interleukin 6(IL-6) were assayed at baseline, 3, 6, and 12-months. The primary endpoint, analysed per-protocol, was change from baseline at 12-months.

Results

Of 52 recruited patients, 24 ablation and 25 rate-control subjects were followed to 12 months. After 1.2 ± 0.5 procedures, sinus rhythm was present in 22(92%) ablation patients; under rate-control, rate criteria were achieved in 23(96%) of 24 patients remaining in AF. At 12 months, MR-proANP fell significantly in the ablation arm(-106.0pmol/L , IQR -228.2 to -60.6) compared with rate-control(-28.7pmol/L , -69 to $+9.5$, $p=0.028$). BNP showed a similar trend towards reduction ($p=0.051$), with no significant difference in Apelin($p=0.13$) or IL-6($p=0.68$). Changes in MR-proANP and BNP correlated with peakVO₂ and ejection fraction, and MR-proANP additionally with quality-of-life score.

Conclusions

Catheter ablation, compared with rate-control, in patients with heart failure and persistent AF was associated with significant reduction in MR-proANP, which correlated with physiological and symptomatic improvement. Ablation-based rhythm-control may induce beneficial cardiac remodelling, unrelated to changes in

inflammatory state. This may have prognostic implications, which require confirmation by event-endpoint studies.

Key Words

Atrial fibrillation; biomarkers; catheter ablation; heart failure; rate-control

Introduction

The optimal therapy for AF in heart failure remains unclear: drug-based 'rhythm-control' has been shown to have no clear advantage over drug-based 'rate-control'.¹ Radiofrequency catheter ablation has been shown to be successful in restoring sinus rhythm in patients with heart failure and to confer clinical benefits.^{2,3}

Biomarkers, including the now well-established use of B-type natriuretic peptide (BNP)^{4,5} and more recently mid-regional pro-atrial natriuretic peptide (MR-proANP)⁶, have become key elements in the diagnosis of heart failure and are known to be strong predictors of morbidity (heart failure-hospitalisation or functional deterioration) and mortality, in turn acting as potential surrogate markers of prognosis. In addition to BNP and MR-proANP, Apelin^{7,8} and the inflammatory marker IL-6^{9,10} may provide additional information on the progress of the heart failure syndrome. The effect of a rhythm-control versus rate-control strategy on these biomarkers, in the context of persistent atrial fibrillation and heart failure, is not clear.

In the recently reported ARC-HF study, a randomized clinical trial comparing catheter ablation with rate control for management of persistent atrial fibrillation in heart failure, we showed a beneficial impact of an ablation strategy upon cardiopulmonary exercise performance.¹¹ In this pre-defined per-protocol analysis, we sought to examine the relative impact of ablation and rate-control on cardiac remodelling as evidenced by changes in MR-

proANP, BNP, and Apelin, and whether there was an interaction with changes in inflammatory state, assessed by parallel measurement of IL-6.

Methods

Study population

Serial blood samples were acquired from patients enrolled in a randomized trial, ARC-HF (catheter Ablation versus Rate Control for management of persistent atrial fibrillation in Heart Failure, ClinicalTrials.gov NCT00878384)¹¹ which tested the hypothesis that ablation-based rhythm-control improves outcomes compared to a conventional rate-control strategy, using exercise, questionnaire and image-based assessment of cardiovascular function as outcomes. The study complied with the Declaration of Helsinki, was approved by the local research ethics committee, and informed consent was obtained from all subjects.

The trial population consisted of patients with persistent (documented AF >7 days in duration) or permanent (persistent AF >1 year) atrial fibrillation and chronic heart failure NYHA class $\geq$ II; LVEF $\leq$ 35 %. The strategies of rate control (target heart rate $\leq$ 80bpm at rest, and $\leq$ 110bpm at 6 minute walk) and catheter ablation were compared in a 1:1 randomised fashion, with primary endpoint of peak oxygen consumption at exercise testing, and secondary endpoints including ejection fraction, 6 minute walk, and Minnesota quality of life score.

Sample acquisition

Prior to exercise testing, peripheral venous blood was collected from patients in the resting state, into tubes containing 7ml ethylene-diamine-tetra-acetic acid. Centrifugation was performed at 1000G for 5 minutes, within 1 hour of sample acquisition, and plasma was extracted via manual pipette into cryo-tubes for freezer storage of 2ml plasma aliquots at -70

degrees C. Samples were acquired at enrolment into the trial, and at subsequent follow-up visits at 3, 6 and 12 months. Samples were thawed once and aliquots taken for all biomarker analyses.

Laboratory assays and reference ranges

Laboratory analyses were performed blinded to the randomisation group, using coded pseudo-anonymised labelling of samples. *MR-proANP* was measured by immunofluorescent assay on the Brahms Kryptor analyser (Thermo scientific, BRAHMS GmbH, Henningsdorf, Berlin, Germany; analytical range 2.1-10000 pmol/L). Previous studies have identified MR-proANP levels in healthy individuals typically <100pmol/L,¹² 120-130pmol/L as a cut-off for the diagnosis of acute heart failure,^{13,14} and >300pmol/L as an independent prognostic marker in chronic heart failure.⁶ *BNP* was measured by immunoassay on the Beckman Access 2 Immunoassay analyser (Beckman Coulter, High Wycombe, UK) using the Alere Triage BNP reagents (analytical range 25-5000pg/ml). This and similar BNP assays have a typical cut-off of 100pg/ml for acute heart failure diagnosis, with levels >300-400pg/ml in significant heart failure and associated with worse prognostic outcome.^{15,16} *Apelin* was measured by ELISA (Phoenix Europe GmbH, Karlsruhe, Germany); an extraction-free protocol was followed. The antibody used in this apelin assay cross-reacts 100% with Apelin-12, 13 and 36. The assay therefore includes all of the above peptides if present in the plasma (analytical range 0.01-100 ng/ml). Typical range in normal controls has been reported 2000-4000pg/ml.^{8,17} *IL-6* was measured by immunoassay on the Beckman Access 2 Immunoassay analyser (Beckman Coulter, High Wycombe, UK; analytical range 2.5-1500 pg/ml, reference range <6.4pg/mL).

Statistical analysis

In order to examine the impact of undergoing ablation, a *per-protocol analysis* was used, with censorship of results after any breach of protocol. The sample size was calculated for the primary endpoint of the pre-existing clinical trial, requiring 25 patients in each group to detect a 2 ± 2.5 ml/kg/min change in peak oxygen consumption at exercise testing.¹¹ The magnitude of possible effect on Apelin and MR-ANP was unknown, however previous observational studies have examined changes in BNP after ablation in similar size populations.^{18,19} Assuming similar variability, our study population would be sufficient to detect a 20 pmol/L reduction in BNP at 80% power, two-sided alpha 0.05.

Baseline values are expressed as mean (SD) for parametric data, median and interquartile range (IQR = 25th to 75th percentiles) for non-parametric data, or frequency/percentage as appropriate. The primary endpoint was change in biomarker plasma level at 12 months, and secondary endpoints were change at 3 and 6 months. Change was assessed by independent group comparison of individual absolute differences from baseline. Data were assessed for normality by histogram, skewness, and Kruskal-Wallis test within SPSS version 20 (IBM). Based upon this, normally distributed data were analysed by independent (two-sample) t-test (adjusted for presence/absence of equal variances by Levene's test) and presented as mean $\pm$ SD, whilst non-parametric data were analysed by Mann Whitney U (Wilcoxon rank-sum) test and summarised as median (IQR). Paired t or Wilcoxon tests were used to analyse individual responses. Correlation analyses were performed using Pearson (parametric) or Spearman Rank (non-parametric) methods. Two-sided P values of less than 0.05 were regarded as statistically significant, and a p-value less than 0.1 was used as an indication of a non-significant trend.

Results

A flow-chart illustrating the study protocol and follow-up is shown in Figure 1. A total of 52 patients were enrolled in the ARC-HF study. One patient withdrew consent for ablation and continued on normal medical therapy, and was excluded from the per-protocol analysis. Two patients had baseline and 3 month assays only: one entered palliative care for end stage heart failure post 3 month follow-up, was unable to attend 6 month follow-up and died prior to the final 12 month follow-up (biomarker results at these time-points also not being available in the original intention-to-treat analysis); another patient requested catheter ablation at 4 months and results were censored after 3 month follow-up. Consequently at 12 months, 25 rate control patients and 24 ablation patients were included for analysis, reduced by 2 patients compared with the equivalent analysis in the original study. Baseline characteristics for the analysed cohort are shown in Table 1. The groups were well-matched for age, gender, heart failure aetiology and clinical characteristics.

After a mean of 1.2 ± 0.5 ablation procedures, sinus rhythm was present in 22 out of 24 (92%) patients at 12-month follow-up in the ablation arm. In the rate-control arm rate-control criteria were achieved in 23 out of 24 (96%) of rate-control patients that remained in AF (1 was cardioverted to sinus rhythm by an ICD shock prior to final follow-up). Resting heart rate was not significantly different between groups at baseline (Table 1), nor at 12 months (ablation 68 ± 7 bpm, rate control 72 ± 14 bpm, $p=0.24$).

Mid-regional atrial natriuretic peptide (MR-proANP)

Baseline values (Table 1) for both MR-proANP and BNP were increased above reference range, similar to prior studies of patients with NYHA II-III heart failure.^{6,16} In the ablation arm, there was a median change of -79.6 pmol/L (IQR -186.2 to -12.7) at 3 months, -123.1 pmol/L (-231.1 to -13.6) at 6 months, and -106.0 pmol/L (-228.2 to -60.6) at 12 months. In

the rate-control arm, there was median change of -39.6 pmol/L (IQR -77.4 to -0.6) at 3 months, -31.1 (-112.9 to +0.3) at 6 months, and -28.7 (-69 to +9.5) at 12 months. In comparison with rate control, the reductions in the ablation arm were non-significant at 3 months (0.277) and 6 months (0.119) but were significant by 12 months ($p=0.028$) (Figure 2).

B-type natriuretic peptide (BNP)

Similarly BNP reduced over time in both catheter ablation and rate-control arms. After catheter ablation, BNP showed a change of -90 pg/ml (IQR -270 to +8) at 3 months, -112 pg/ml (-380 to -3) at 6 months, and -120 pg/ml (-285 to +8) at 12 months. Under rate control, the changes were -26 pg/ml (IQR -74 to +22) at 3 months, -24 pg/ml (-84 to +56) at 6 months, and -16 pg/ml (-88 to +36) at 12 months. Comparing the groups, the reduction in BNP in the ablation arm was not significant at 3 months ($p=0.17$) however showed a strong non-significant trend towards greater reduction post-ablation at 6 ($p=0.052$) and 12 months ($p=0.051$). The sequential change for plasma BNP is presented in Figure 3. It is noted that, under the original intention-to-treat protocol of the ARC-HF trial, the outcome for BNP did reach significance for a reduction with ablation over rate-control ($p=0.038$ at 6 months and $p=0.045$ at 12 months)¹¹: the difference may reflect borderline power of the sample size, noting that the per-protocol analysed cohort was reduced by 2 patients at 6 and 12 months in comparison with the original.

Apelin

Baseline Apelin levels were lower than typical reference ranges, consistent with earlier data.⁸ In the ablation arm, median change was -27 pg/ml (IQR -250.25 to +161.5) at 3 months, -89.5 pg/ml (-238.5 to +52.5) at 6 months, and +9.5 pg/ml (-153.5 to +139.75) at 12 months. By comparison, in the rate control arm, median change was +74.5 (IQR -65 to 205.25) at 3 months, +133 pg/ml (-40 to +240) at 6 months, and +120 pg/ml (-9 to +294) at 12 months.

The difference in groups was non-significant at 3 months ($p=0.22$), although there was further change reaching significance at 6 months ($p=0.002$). However, following an overall increase in Apelin level in the final 6 months, comparison at 1 year was non-significant ($p=0.134$). The sequential changes in plasma apelin are displayed graphically in Figure 4.

Interleukin-6 (IL-6)

Overall, average IL-6 levels around the upper reference value at baseline. In the catheter ablation group, IL-6 showed a median change of $+0.07$ pg/mL (IQR -0.45 to $+0.76$) at 3 months, $+0.06$ pg/mL (IQR -0.52 to $+1.38$) at 6 months, and -0.06 pg/mL (IQR -0.64 to $+1.69$) at 12 months. In the rate control group, the change was -0.1 pg/mL (IQR -0.89 to 0.51) at 3 months, $+0.11$ pg/mL (IQR -0.78 to $+0.88$) at 6 months, and $+0.34$ pg/mL (IQR -0.47 to $+0.87$) at 12 months. Comparing the groups, there was no significant difference at 3 ($p=0.93$), 6 ($p=0.32$) or 12 months ($p=0.68$). Results for change in IL-6 from baseline are shown graphically in Figure 5.

Correlation of biomarkers with ARC-HF endpoints

The trends in change for MR-proANP and BNP following ablation, versus rate-control, were concordant with the change in peak VO₂, quality of life score and left ventricular ejection fraction found in the ARC-HF trial.¹¹ Correlation analysis (Table 2) showed that MR-proANP correlated best with all 3 endpoints, and most closely correlated with peak VO₂, whilst BNP correlated well with peak VO₂ and better than the other biomarkers with change in LVEF. Change in both biomarkers correlated moderately with change in left atrial size. MR-proANP and BNP correlated well together (change at 12 months $R=0.63$, $p<0.001$). There were no significant correlations between Apelin or IL-6 and any of the original endpoints.

Discussion

This study investigated the impact of catheter ablation-based rhythm control versus medical rate-control on cardiac and inflammatory biomarkers in patients with persistent AF and systolic heart failure. At 12 months, the resulting prevalence of sinus rhythm was nearly as high in the ablation group (92%) as that of rate-controlled AF in the rate-control group (96%). Hence the study examines not only strategy but also, effectively, the effect of sinus rhythm versus AF at follow-up.

The most significant impact was on MR-proANP, which showed an overall trend towards reduction in both groups, with significantly greater reduction in the ablation arm at 12 months. Whilst, atrial natriuretic peptide (ANP) proved to be inferior to BNP in terms of its diagnostic and prognostic potential, largely due to lack of reproducibility, the recently introduced assay of the mid-regional segment of its precursor, MR-proANP has recently been identified as an important biomarker, with several studies indicating its potential role as a predictor of morbidity and mortality.^{6,20} It has been shown to have similar accuracy to BNP as a diagnostic test,⁶ and may be a stronger prognostic marker.²⁰ Additionally it may be superior to BNP for detecting and managing high-risk patients with chronic heart failure.¹⁶ Increases of $\geq 30\%$ have appeared to define patients at highest risk of events, although more modest changes also have an impact.¹⁶ In the current study, the median reduction in MR-proANP after ablation was 28%, compared with 7% for rate control at 12 months. Although in the current study, the potential prognostic implications are purely speculative, the finding that changes in MR-proANP correlate strongly with changes in VO_2 (and moderately well with changes in LVEF and quality of life score) would further support the emerging evidence supporting use of this biomarker in assessing the impact of treatment and assisting risk stratification of patients with heart failure.¹⁶ The significant reduction in MR-pro-ANP over

time may also suggest a favourable effect on atrial remodelling in addition to the presumed beneficial effects on LV structure and function.

Although it has been shown that MR-proANP levels are higher in non-heart failure patients with AF compared with those in normal sinus rhythm,²¹ and the change of rhythm itself might be regarded as one explanation for the observed fall and thus be a potential confounder, it should be noted that similar values were observed in those with heart failure regardless of rhythm²¹ and – like peak VO_2 in the main ARC-HF trial¹¹ – the change was not entirely explained by restoration of rhythm alone, being more marked at (6 and) 12 months than at 3 months despite the same overall rate of sinus rhythm maintenance. Therefore, although the early change may reflect a combination of rhythm-change and cardiac remodelling, later differences are unlikely to be explained by analysis of patients in different rhythms alone, and suggest progressive regression of the heart failure syndrome after restoration of sinus rhythm.

It has been shown that BNP and N-terminal pro-BNP levels are not independently affected by AF in advanced heart failure,^{22,23} thus BNP is well placed for assessing serial change in cardiac function. Notably, BNP levels have been shown to fall after catheter ablation for paroxysmal and persistent AF in patients without ventricular dysfunction.^{18,19} In the per-protocol analysis used in this biomarker study, the reduction in BNP showed a trend rather than reaching statistical significance as per the original intention-to-treat analysis,¹¹ probably due to the effect of drop out. However, the changes in BNP observed in the present study showed similar trends to MR-proANP, and indeed the two measures were highly correlated. Furthermore, change in BNP correlated better than MR-proANP with change in LVEF, as expected given its relationship with ventricular strain and failure.

IL-6, an inflammatory cytokine, has been shown to increase in patients with AF,^{24,25} may contribute to its pro-thrombotic state,²⁶ and is increased acutely after ablation.²⁷ IL-6 is also known to be raised in patients with chronic heart failure,¹⁰ and to be an independent predictor of mortality and worsening heart failure.^{28,29} Previous studies have shown a potential link between changes in inflammatory state and that of cardiac biomarkers including BNP,³⁰ and there is limited evidence for IL-6 mediating upregulation of BNP during cardiac stress.³¹ In the present study, IL-6 did not show a significant elevation at baseline, change over time, or difference between groups at follow-up, and these findings suggest i) that ablation, beyond the acute post-operative period, has an apparently neutral effect on markers of inflammation in the context of pre-existing persistent AF and heart failure, and ii) that the changes in MR-proANP and BNP are independent of a change in IL-6 – which may be reflective of the inflammatory state.

The relatively novel peptide *apelin* is an endogenous ligand with potent inotropic effects.³²⁻³⁴ Apelin levels are known to be reduced in patients with chronic heart failure when compared with controls⁸, and are raised by intervention such as biventricular pacing,³⁵ although other studies have shown that levels may be increased in mild-moderate heart failure, and decrease back towards control levels in severe heart failure.⁷ Apelin, perhaps as a consequence of this heterogeneity, is the most difficult of the biomarkers to interpret, both in terms of the results of this study and their clinical and pathophysiological relevance. In the presence of the latter pattern of behaviour, varied response to restoration of improved haemodynamics might be expected, with patients starting from different positions on the scale, and may be one explanation for the lack of a clear difference between Apelin levels in the two treatment arms. Furthermore, plasma Apelin itself may be an insufficient marker in isolation, without

considering its binding interaction with the APJ receptor at the cardiac level, which appears to be down-regulated in heart failure and up-regulated during reverse ventricular remodelling.⁷

Study Limitations

This study used a per-protocol analysis, which may overestimate clinical effects from treatment strategies, although this protocol was pre-specified and sought to examine the real impact of catheter ablation rather than using an intention-to-treat analysis as was appropriate for the principal clinical trial outcomes. None of the biomarker data-sets were normally distributed, at least in part due the relatively small sample size, and therefore non-parametric testing was used: this limits conclusive comments to be made about the magnitude of change when comparing the treatment arms and precluded serial measures analysis by summary measures, including area-under-curve³⁶ which is not pragmatically feasible without application of parametric testing. Analysis of change at 3 and 6 months (secondary endpoints) did not adjust for multiple comparisons and should be considered exploratory.

Conclusion

This study has shown, for the first time, that a catheter-ablation based rhythm control strategy, when compared with rate-control, significantly lowers MR-proANP at follow-up. BNP showed a similar trend, whilst there was no overall effect on Apelin or IL-6. The reduction in MR-proANP, and to a lesser extent BNP, correlated with improved cardiopulmonary, quality-of-life and imaging indices. Overall, these findings would suggest a beneficial effect of a catheter ablation strategy, unrelated to any demonstrable changes in the inflammatory state, with possible prognostic relevance that will require further evaluation in event-endpoint driven clinical trials.

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Author Contributions

D G Jones - conception and design of the study, acquisition of data, analysis and interpretation of data, statistical analysis, obtaining funding, drafting of the manuscript, critical revision of article, final approval of the manuscript; S K Haldar - acquisition of data, critical revision of article, final approval; J Donovan - acquisition of data, analysis and interpretation of the data, critical revision of article, final approval; T A McDonagh - conception and design of the research, critical revision of article, final approval; R Sharma - conception and design of the research, critical revision of the article, final approval; W Hussain - drafting of the manuscript, critical revision of the article, final approval; V Markides - conception and design of the research, obtaining funding, critical revision of the article, supervision, final approval; T Wong - conception and design of the research, obtaining funding, drafting of the manuscript, critical revision of the article, supervision, final approval

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Figure 1 ARCHF-biomarker study flowchart.

Number of patients included for per-protocol analysis shown at each time point

Figure 2 Mid-Regional proAtrial Natriuretic Peptide (MR-proANP)

Change (Δ) in mid-regional proANP (MR-proANP) from baseline at follow-up, represented as median with error bars showing 25th and 75th percentiles (IQR): catheter ablation (solid line) vs. rate control (dashed line). Statistical significance* is shown at 12 months between groups ($p=0.028$)

Figure 3 B-type Natriuretic Peptide (BNP)

Change (Δ) in BNP from baseline at follow-up time points, represented as median with error bars showing IQR: catheter ablation (solid line) vs. rate control (dashed line). The change at 6 ($p=0.052$) and 12 months (0.051) was borderline, showing a statistical trend[^] rather than meeting significance criteria.

Figure 4 Apelin

Change (Δ) in Apelin from baseline at follow-up time points, represented as median with error bars showing IQR: catheter ablation (solid line) vs. rate control (dashed line). ** indicates $p=0.002$ at 6 months, whereas the difference between groups was not statistically significant at 12 months

Figure 5 Interleukin-6 (IL-6)

Change (Δ) in IL-6 from baseline at follow-up time points, represented as median with error bars showing IQR: catheter ablation (solid line) vs. rate control (dashed line).

Table 1 Baseline characteristics

	Rate control n=25 N (%) / mean±SD	Catheter Ablation n=24 N (%) / mean±SD	p value
Age	62±9	64±10	0.56
Male	23 (92)	19 (79)	0.19
Ischaemic HF aetiology	6 (24)	10 (42)	0.19
Time since HF diagnosis (months)	49±58	63±62	0.40
Time since AF diagnosis (months)	53±77	49±37	0.84
Duration of continuous AF (months)	24±30	23±22	0.81
NYHA class	2.5±0.5	2.5±0.5	0.88
<i>NYHA II</i>	<i>13 (52)</i>	<i>13 (54)</i>	
<i>NYHA III</i>	<i>12 (48)</i>	<i>11 (46)</i>	
Recent HF hospitalisation (<1year)	7 (28)	8 (33)	0.69
Minnesota LHFQ score (/105)	49±21	42±22	0.26
Creatinine (μmol/L)	103±28	95±23	0.30
Peak VO ₂ (ml/kg/min)	18.3±4.9	16.5±5.5	0.23

QRS duration (ms) <i>non-paced only</i>	114±21	119±19	0.45
Radionuclide LVEF (%)	25±7	22±8	0.21
LA area (apical 4-chamber, cm ²)	27±7	30±6	0.15
6MWD (m)	412±111	416±81	0.88
Resting HR (bpm)	81±12	78±9	0.31
Exercise HR (bpm)	109±18	109±16	0.94
Proportion rate-controlled at baseline (≤80 rest, ≤110 at 6MW)	14 (56)	15 (62)	0.64
Beta blocker	23 (92)	22 (92)	0.63
ACE or ARB	25 (100)	23 (96)	0.49
Aldosterone antagonist	5 (20)	12 (50)	0.04
Amiodarone	3 (12)	3 (12)	1.0
Digoxin	12 (48)	14 (58)	0.47
Preexisting ICD; of which CRT	4 (15); 3 (12)	9 (38); 7 (31)	0.08 ; 0.13
Ventricular pacing dependent	1 (4)	1 (4)	0.51
<i>Biomarker baseline values</i>			
Apelin (pg/ml)	867±425	813±289	0.61
BNP (pg/ml)	288±290	386±301	0.25

IL-6 (pg/ml)	4.5±3.1	5.3±6.9	0.59
MR-proANP (pmol/L)	355±199	367±147	0.81

ACE = angiotensin converting enzyme inhibitor, ARB = angiotensin receptor blocker, CRP = C-reactive protein, CRT = cardiac resynchronization therapy, HF = heart failure, HR = heart rate, ICD = implantable cardioverter defibrillator, LHFQ = Minnesota Living with Heart Failure Questionnaire, LVEF = left ventricular ejection fraction, NYHA = New York Heart Association, 6MWD = 6-minute walk distance.

Table 2 Correlation of biomarkers with ARC-HF endpoints

	ΔMRANP		ΔApelin		ΔBNP		ΔIL-6	
	n=49	<i>p</i>	n=49	<i>p</i>	n=48	<i>p</i>	n=49	<i>p</i>
ΔVO₂	-0.68	<0.001	0.13	0.36	-0.63	<0.001	0.01*	0.93
ΔLHFQ	0.35	0.015	0.14	0.30	0.22	0.14	-0.04	0.77
ΔLVEF	-0.32	0.025	-0.16	0.28	-0.53	<0.001	0.18*	0.23
ΔLAa	0.42	0.003	0.04	0.77	0.43	0.002	0.01*	0.93

For each biomarker correlation estimations were performed, using Spearman Rank (or *Pearson) correlation, against change (Δ) from baseline to 12 months follow-up, based on the per-protocol analysis (see Methods). Correlation coefficient values (R) are shown with corresponding 2-sided p-values; n = number of patients samples available for analysis

BNP = B-type natriuretic peptide, IL-6 – Interleukin-6, LAa = left atrial area (apical 4 chamber) LHFQ = Minnesota Living with Heart Failure Questionnaire score, LVEF = left ventricular ejection fraction, MRANP = midregional pro-atrial natriuretic peptide, VO2 = peak oxygen consumption at cardiopulmonary exercise test