Journal of Cardiac Failure Vol. 18 No. 10 2012

Genetic Variants of the Renin-Angiotensin-Aldosterone System and Reverse Remodeling After Cardiac Resynchronization Therapy

RENATA DE MARIA, MD,[1] MAURIZIO LANDOLINA, MD,[2] MAURIZIO GASPARINI, MD,[3] BORIS SCHMITZ, PhD,[4] JONICA CAMPOLO, MSc,[1] MARINA PAROLINI, BStat,[1] ANTONIO SANZO, MD,[2] PAOLA GALIMBERTI, MD,[3] MICHELE BIANCHI, MD,[5] STEFAN-MARTIN BRAND, MD, PhD,[4,6] OBERDAN PARODI, MD,[1] AND MAURIZIO LUNATI, MD[5]

Milan, Pavia, and Rozzano-Milan, Italy; and Münster, Germany

ABSTRACT

Background: Reverse remodeling (RR) after cardiac resynchronization therapy (CRT) is associated with favorable clinical outcomes in heart failure (HF). The renin-angiotensin-aldosterone system (RAAS) is involved in the remodeling process.

Methods and Results: We assessed the association between RR and 8 common RAAS gene variants, which were determined by TaqMan assays, in 156 outpatients with chronic HF. RR was defined as a >15% decrease in left ventricular end systolic volume (LVESV) at 9 (interquartile range 7−12) months after CRT. We matched 76 patients who did not show RR (RR−) to 80 RR+ control subjects by age, sex, HF etiology, New York Heart Association (NYHA) functional class and left ventricular ejection fraction (LVEF). The frequency of the minor allele of the *NR3C2* gene (rs5522 C/T), encoding the mineralocorticoid receptor, was higher in RR− than in RR (24/126 vs 10/150; *P* value after false discovery rate correction: <.0193). Conversely, LVESV decreased significantly less after CRT in carriers of the *NR3C2* minor C allele ($P = .02$). After adjustment for age, sex, NYHA functional class, previous myocardial infarction, atrial fibrillation, and LVEF, RR− remained independently associated with *NR3C2* C allele carriage (odds ratio 3.093, 95% confidence interval 1.253−7.632).

Conclusions: The association of RR− after CRT with a common polymorphism in the mineralocorticoid receptor gene involved in aldosterone signaling suggests a possible role for variants in RAAS genes in progressive LV function decline, despite apparently effective CRT. (*J Cardiac Fail 2012;18:762−768*)

Key Words: Heart failure, cardiac resynchronization therapy, reverse remodeling, NR3C2, mineralocorticoid receptor.

Reverse remodeling (RR) of dilated failing ventricles is a generally accepted goal in the treatment of heart failure (HF). RR can be obtained by medical therapy, as demonstrated by the landmark randomized controlled trials of renin-angiotensin-aldosterone system (RAAS) inhibitors and beta-blockers. More recently, cardiac resynchronization therapy (CRT) has also been shown to trigger RR in patients with advanced HF and dyssynchrony, even when on optimal medical treatment. Besides improving clinical symptoms, quality of life, exercise tolerance, and survival in patients with advanced HF,[1,2] CRT reverses the remodeling process by reducing ventricular size, mass, and mitral regurgitation in both short and long terms.[3−5]

RR, as defined by decreases in left ventricular end-systolic volume (LVESV) compared with preprocedural values, peaks on average within 6 months after CRT and has been associated

From the [1]CNR Institute of Clinical Physiology, Cardiothoracic and Vascular Department, Niguarda Ca' Granda Hospital, Milan, Italy; [2]Department of Cardiology, Fondazione IRCCS Policlinico San Matteo, Pavia, Italy; [3]Department of Cardiology, Istituto Clinico Humanitas IRCCS, Rozzano-Milan, Italy; [4]Medical Faculty of the Westfalian Wilhelms−University of Münster, Department of Molecular Genetics of Cardiovascular Disease, Münster, Germany; [5]Cardiovascular Department, Niguarda Ca' Granda Hospital, Milan, Italy and [6]Leibniz-Institute for Arteriosclerosis Research, Münster, Germany.

Manuscript received March 6, 2012; revised manuscript received July 27, 2012; revised manuscript accepted July 31, 2012.

Reprint requests: Renata de Maria, MD, CNR Institute of Clinical Physiology, Cardiothoracic and Vascular Department, Niguarda Ca' Granda Hospital, Piazza Ospedale Maggiore 3, 20162 Milan, Italy. Tel: +39 026473407; Fax: +39 0266116990. E-mail: renata_de_maria@hotmail.com

Funding: Partially supported by the European Community Seventh Framework Programme ICT 2007 project VPH2 (grant agreement no. 224635).

See page 767 for disclosure information.

1071-9164/$ - see front matter

http://dx.doi.org/10.1016/j.cardfail.2012.07.008

with better clinical outcomes.[3,6–8] Failure of RR after CRT is therefore an important clinical issue with relevant prognostic implications. Demographic, clinical, and procedural variables have been associated with RR after CRT,[9–15] but no data are available currently on the possible association of genetic variants with an RR phenotype.

The RAAS plays a major role in the pathophysiology of HF, through increased vasoconstriction, sodium and water retention, myocardial fibrosis, and ventricular remodeling, and is a generally accepted therapeutic target in HF. Using the candidate gene approach, genetic variance in the RAAS has been previously associated with left-ventricular hypertrophy, remodeling or HF (susceptibility genes), or other specific phenotypic expressions (modifier genes).[16,17]

We assessed, using a multicenter case-control design, the prevalence of genetic variants in 6 RAAS genes that play pivotal roles in cardiovascular function or signal transduction and that most likely affect the RR process in patients with chronic systolic HF and different degrees of reverse left ventricular (LV) remodeling after CRT.

Methods

Study Population

Out of a total population of 1,421 patients implanted with CRT since 2002 at the 3 participating institutions, we enrolled 160 consenting subject who had undergone CRT ≥12 months earlier and were consecutively reviewed in the electrophysiology outpatient clinics of the 3 participating institutions from March to December 2009. CRT had been performed in all according to current guidelines,[18] ie, advanced heart failure (New York Heart Association [NYHA] functional class II–IV), wide QRS complex (≥120 ms), depressed LV function (ejection fraction [LVEF] ≤35%), and LV end-diastolic diameter >55 mm at baseline, through transvenous implantation of a right ventricular defibrillation lead, an atrial pacing lead (except for patients in atrial fibrillation), and a coronary sinus lead for LV pacing. Entry criteria also included stable positioning of the left lead at the lateral or posterolateral wall level and ventricular volumes measured at follow-up echocardiography 6–12 months after CRT. Patients with procedural failure, particularly a nonfunctioning LV lead, were excluded.

Lack of reverse remodeling (RR−) was defined as any change in LVESV at follow-up echocardiography, 6–12 months after CRT >−15% compared with baseline values. For each RR− patient, a control subject with LVESV decrease >15% (RR+) matched by sex, NYHA functional class, HF etiology, age (within 10 years), and baseline LVEF (within 5%) was enrolled.

Blood was sampled for genetic analysis during a follow-up outpatient visit. Timing of data collection was baseline, ie, the last available data before CRT, and follow-up, ie, 6–12 months after the procedure.

The study was approved by the Institutional Ethics Committees of the 3 participating centers. Patients expressed their written informed consents to participate.

Genotyping

The selected common variants within 6 RAAS genes have been replicatively associated with HF phenotypes[16] and were considered as markers and/or functional variants. For the *ACE* gene, we included 3 variants, because linkage and association analysis suggested their combinations to be better predictors for angiotensin-converting enzyme (ACE) protein levels than either one alone.[19] We included only variants with previously reported minor allele frequencies of >10% in a European ancestry population.

Genomic DNA was extracted from 200 μL of patients' EDTA blood with the use of the QIAamp DNA Blood kit (Qiagen, Hilden, Germany) according to the manufacturer's instructions. Genotyping was performed at the University Clinic of Münster, Germany, blinded to remodeling status, with the use of TaqMan SNP Genotyping Assays. Assays were purchased from Applied Biosystems (Life Technologies Corporation, Carlsbad, California, USA) and were applied using the ABI7900 real-time polymerase chain reaction (PCR) system. Real-time PCR was performed in a 384-well format with the use of 2.5 μL TaqMan Genotyping Master Mix (2×), 0.125 μL TaqMan SNP Genotyping Assay (40×), 2.375 μL DNase-free water, and 2 ng DNA. Real-time PCR conditions were as follows: initial denaturing at 95°C for 10 minutes, 40 cycles of 95°C for 15 seconds, and 60°C for 1 minute. Replicate samples and samples without template were used as control.

From the original population of 160 subjects from whom blood was sampled, successful genotyping was available for 156 patients.

Echocardiographic Measurements

All patients underwent a comprehensive transthoracic echocardiographic examination at rest, with the use of conventional methods using commercially available ultrasound machines equipped with a 2.5–3.5-MHz phased-array sector scan probe, second harmonic technology. LV volumes were measured and LVEF obtained by 2- and 4-chamber view (Simpson's rule). Intra- and interoperator variabilities for volume measurements were 5% and 6%, respectively.

Statistical Analysis

Data are expressed as median and interquartile range and frequency (%). Chi-square test was used to compare categoric variables, or the Fisher exact test if necessary, with Mantel-Haenszel linear-by-linear association where appropriate. Student *t* test or Mann-Whitney test was used to compare continuous variables. The correlation between changes in LVESV and LVEF was investigated by Spearman correlation and presented by ρ and *P* values. Concordance between RR criteria derived from LVESV or LVEF changes was assessed by the κ statistic.

Genotyping call rates were all >98%. Hardy-Weinberg equilibrium was tested by calculating the expected genotype frequencies from the allele frequencies; deviation from the observed genotype frequencies was determined by chi-square. Genotype distributions in the 6 RAAS pathway genes analyzed were compatible with Hardy-Weinberg equilibrium, except for rs1403543 polymorphism in the *AGTR2* (angiotensin receptor 2) gene, which was consequently dropped from further analysis, leaving 5 genes and 7 SNPs to be tested for association with RR (Table 1). Genetic association was tested by comparing allelic frequencies between RR− and RR+ groups. To correct for multiple comparisons, we used the Benjamini and Yekutieli false discovery rate method.[20] Statistical significance was declared at $P \leq .0193$ using the formula $P = a/\Sigma(1/i)$, where $a = .05$ and i ranges from 1 to N, where $N = 7$ represents the number of allele comparisons.

Group sample sizes of 76 RR− and 80 RR+ achieve >80% power to detect an allele frequency difference of 0.10 (odds ratio [OR] 2.58) for an allele of 11% population frequency.

Table 1. Association of the Studied Polymorphisms with Cardiovascular Phenotypes

Gene	SNP	Major Allele	Minor Allele	Reported Association
AGT	rs699	G	A	HF occurrence; LV hypertrophy; LV size
ACE	rs4291	A	T	Hypertension
ACE	rs4646994	Ins	Del	HF occurrence and phenotype; LV hypertrophy; LV size
ACE	rs4343	A	G	Reduced risk of hypertension (A allele)
AGTR1	rs5186	A	C	HF phenotype; LV hypertrophy; LV size
CYP11B2	rs1799998	T	C	HF phenotype; LV hypertrophy; LV size
NR3C2	rs5522	T	C	Enhanced physiologic stress responses

ACE, angiotensin-converting enzyme; AGT, angiotensinogen; AGTR1, angiotensin receptor 1; CYP11B2, aldosterone synthase; HF, heart failure; LV, left ventricular; NR3C2, mineralocorticoid receptor; SNP, single-nucleotide polymorphism.

The association between lack of RR and genetic variants was adjusted for clinically relevant potential confounders by multivariable logistic regression. Odds ratios with their corresponding 95% confidence intervals (CIs) were estimated. Hosmer-Lemeshow *U*-statistic was calculated as a measure of fit.

Two-sided *P* values of $<.05$ were considered to be statistically significant. The Statistical Package for the Social Sciences (SPSS) v17 was used for all analyses.

Results

Characteristics of RR

Table 2 shows the preprocedural clinical characteristics of the study population. LVESV changes were assessed at follow-up 9 (7−12) months after CRT, compared with the preprocedural echocardiogram. Eighty patients were classified as RR+ and 76 as RR−. The 2 groups were well matched by baseline clinical characteristics, except for prevalence of diabetes (RR− 24% vs RR+ 11%; $P = .06$).

After CRT, both LV volumes decreased and LVEF improved significantly in RR+, whereas they were unchanged in RR− (Fig. 1). As expected, changes from baseline to follow-up in LVESV and LVEF were correlated (Spearman ρ coefficient = −0.72; $P < .001$). Concordance between the 15% decrease LVESV cutoff and improved LVEF was moderate: κ value was 0.56 ($P < .001$) for an absolute 5% LVEF increase and 0.43 ($P < .001$) for a 15% relative LVEF increase. At follow-up (Table 3), a significantly greater proportion of RR+ than of RR− subjects improved by at least 1 NYHA functional class (75% vs 55%; $P < .01$), whereas no differences were observed in drug treatment between groups.

RAAS Variants and RR

Allele frequencies of the 7 analyzed SNPs are shown in Table 4. Only the minor (C) allele frequency of the *NR3C2* gene, encoding the mineralocorticoid receptor (MR), was significantly higher in RR− than in RR+. After CRT, LVESV decreased significantly less in carriers of *NR3C2* minor allele ($P = .02$; Fig. 2). Conversely, the *NR3C2* minor C allele was associated with a graded lesser extent of RR ($P = .0008$; Fig. 3).

MR antagonist treatment conditions did not differ between carriers of the minor allele and noncarriers: Administration rates were 67% vs 61% ($P = .09$), and daily doses

Table 2. Characteristics of the Study Population Before CRT

	All (n = 156)	RR− (n = 76)	RR+ (n = 80)	*P* Value
Sex, male	136 (87%)	69 (91%)	67 (84%)	.23
Ischemic etiology	79 (51%)	40 (53%)	39 (49%)	.63
History of hypertension	43 (27%)	22 (31%)	21 (28%)	.72
Diabetes	27 (17%)	18 (24%)	9 (11%)	.06
Previous myocardial infarction	63 (41%)	33 (44%)	30 (39%)	.51
Atrial fibrillation	25 (16%)	14 (18%)	11 (14%)	.51
Anemia (Hb <11.5 g/dL)	11 (8.5%)	4 (6.3%)	7 (10.4%)	.53
Kidney dysfunction (creatinine >2 mg/dL)	7 (5.5%)	4 (6.3%)	3 (4.7%)	1.00
NYHA functional class III−IV	111 (71%)	53 (70%)	58 (72%)	.73
Beta-blockers	126 (82%)	63 (83%)	63 (83%)	1.00
RAAS inhibitors	145 (97%)	75 (99%)	70 (96%)	.36
MR antagonists	97 (64%)	51 (67%)	46 (61%)	.50
Age (y)	62 (56−70)	61 (56−70)	64 (57−71)	.68
Months since symptom onset	60 (24−96)	60 (24−114)	48 (24−80)	.25
QRS duration (ms)	160 (140−180)	160 (140−180)	169 (150−188)	.16
LVEF (%)	27 (22−30)	27 (23−30)	27 (22−30)	.66
LVEDV (mL)	227 (190−310)	227 (174−295)	230 (200−330)	.25
LVESV (mL)	170 (135−231)	164 (121−222)	178 (140−240)	.25
Months to follow-up echo after CRT	9 (7−12)	10 (7−12)	9 (7−12)	.88

LVEDV, left ventricular end-diastolic volume; LVEF, left ventricular ejection fraction; LVESV, left ventricular end-systolic volume; MR, mineralocorticoid receptor; NYHA, New York Heart Association; RAAS, renin-angiotensin-aldosterone system; RR, reverse remodeling.

Values are expressed as n (%) or median (interquartile range).

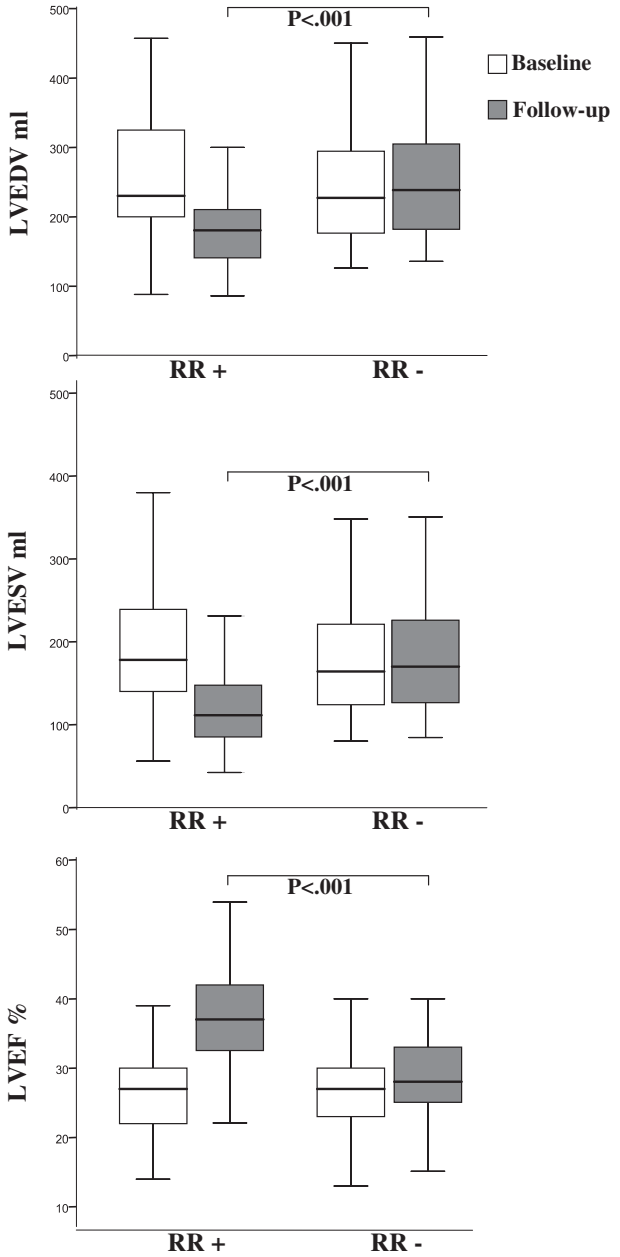

Fig. 1. Left ventricular (LV) end-diastolic (LVEDV) and end-systolic (LVESV) volume and ejection fraction (LVEF) at baseline (open boxes) and follow-up (filled boxes) according to the prespecified reverse remodeling phenotype. Between-group differences at follow-up were significant for all measurements($P < .001$). Box plots show median (line), interquartile range (box), and extreme values (whiskers).

were 25 (25−37) mg vs 37 (25−50) mg ($P = .22$), respectively.

By multivariable logistic regression, after adjustment for age, sex, previous myocardial infarction, NYHA functional class, atrial fibrillation, baseline LVEF, and diabetes, presence of the minor allele in the *NR3C2* gene remained independently associated with lack of RR during follow-up (Table 5). The model showed good calibration (Hosmer-Lemeshow goodness of fit statistic: $P = .95$).

Table 3. Follow-Up Functional Class and Drug Treatment According to Evidence of RR, n (%)

	All (n = 156)	RR− (n = 76)	RR+ (n = 80)	*P* Value
NYHA functional class				.01
Unchanged/increased	54 (35%)	34 (45%)	20 (25%)	
Decreased ≥1	100 (65%)	41 (55%)	59 (75%)	
Beta-blockers	136 (88%)	67 (88%)	69 (89%)	1.00
RAAS inhibitors	144 (97%)	73 (96%)	71 (97%)	1.00
MR antagonists	81 (53%)	43 (57%)	38 (48%)	.34

Abbreviations as in Table 2.

Discussion

To the best of our knowledge, this is the first report on the role of variants in candidate genes linked to cardiac structure and function in LV remodeling response after CRT. The main finding of our study that excluded subjects with procedural failure is the association between lack of RR after CRT and the minor allele of the *NR3C2* gene involved in aldosterone signaling, even after adjustment for known clinical confounders.

RR occurs in more than one-half of patients and peaks between 6 and 12 months after CRT. Its prevalence varies with the echocardiographic variable and cutoff chosen and timing of assessment.[6,7,21,22] We used LVESV decrease as marker of RR because of its reported association with outcomes, and selected the more stringent cutoff of 15%, rather than the 10% criterion coupled to survival at 6 months, because by definition all of our patients were alive at the time of genetic characterization. Consistent with previous findings,[22] we observed only moderate agreement between LVESV and LVEF remodeling cutoff values. However, RR+ showed a significantly greater LVEF improvement than RR− (Fig. 1).

In our white population, which is representative of other series of HF patients on optimized medical treatment undergoing CRT, 51% of the subjects showed a LVESV decrease of >15% at a median time of 9 months after the procedure. The observed balance between RR− and RR+ in acknowledged key clinical predictors of outcome dims their confounding effect and minimizes bias in the evaluation of genetic variance. Factors underlying response to CRT regarding RR are probably many, and among them patients' genetics may well have a role.[23]

RR− patients had a higher prevalence of diabetes of borderline significance. Several reports have focused on the impact of diabetes on response to CRT.[24,25] Intriguingly, all of our diabetic patients carrying the minor allele of the *NR3C2* gene had an RR− phenotype, but the association was not confirmed after adjustment for clinical confounders, including previous myocardial infarction.

Because the scope of our investigation was restricted to patients in whom technically successful CRT was not followed by improved chamber size and pump function, we investigated RAAS genetic variants linked to altered protein function and ventricular structure. In this population, only

Table 4. Allelic Frequencies of the Analyzed RAAS Gene Variants According to RR (n = Number of Successfully Tested Cases)

Gene	SNP	Alleles	RR− (n = 76)	RR+ (n = 80)	*P* Value* (Allele)
AGT	rs699 (n = 155)	A/G	89/63	90/68	NS
ACE	rs4291 (n = 155)	T/A	53/97	56/104	NS
ACE	rs4646994	Ins/Del	65/87	63/97	NS
ACE	rs4343	G/A	82/70	96/64	NS
AGTR1	rs5186 (n = 155)	C/A	49/103	43/115	NS
CYP11B2	rs1799998 (n = 155)	C/T	78/74	64/94	NS
NR3C2	rs5522 (n = 155)	C/T	24/126	10/150	<.0193

Del, deletion; Ins, insertion; other abbreviations as in Tables 1 and 2.
*Significance level after false discovery rate correction: <.0193.

the rs5522 polymorphism of the *NR3C2* gene was significantly associated with lack of RR, suggesting its possible modifier effect on the putative mechanisms of structural change.

The deleterious role of aldosterone and MR activation in the heart has recently been extensively reviewed.[26–29] Aldosterone induces hypertrophy and the dysregulation of proliferation and apoptosis in the myocardium and vessel wall, which lead to fibrosis and chamber remodeling. MR activation, by changes in redox status or by aldosterone levels inappropriate for sodium status, is accompanied by vascular inflammation and end-organ damage. Deletion of MR in cardiomyocytes ameliorates adverse remodeling after myocardial infarction in the rodent model.[30] In clinical trials in both advanced and mild HF,[31] MR antagonists at doses devoid of relevant natriuretic effects improved mortality and morbidity, despite plasma aldosterone levels in the low to normal range. The adverse effects of MR stimulation may in fact be independent from peripheral hormone concentrations. In the Framingham Offspring Study,[32] there was no association between plasma aldosterone levels and variants in the MR and aldosterone synthase genes, with an estimated heritability of serum aldosterone of 0.10. In the RALES (Randomized Aldactone Evaluation Study) trial, 25 mg spironolactone had no apparent effect on sodium retention score, urinary sodium excretion, or body weight but was associated with reduced mortality and morbidity.

The negative effects of aldosterone on myocardial tissue might be mediated by genetically modulated increased activity of the MR receptor. The independent association between *NR3C2* minor allele and RR− phenotype in the present population suggests that this genetic variant might result in MR up-regulation, heightened susceptibility even to normal aldosterone levels, adverse changes in extracellular matrix, and a lower probability of favorable CRT effects.

Study Limitations

The population studied in this pilot investigation, although phenotypically well characterized and balanced across many clinical confounders, was retrospectively enrolled, relatively small, and possibly not adequately powered to detect even small phenotypic differences for all genetic variants studied. However, we used stringent phenotypic criteria for RR that had previously been validated through hard end points of morbidity and mortality[6–8] and restricted our analysis to genes with reported minor allele frequencies of >10% in a population with European ancestry.

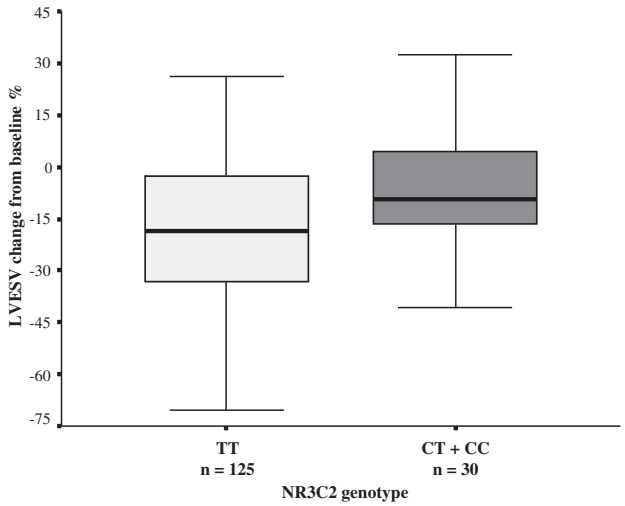

Fig. 2. Distribution of percentage changes in left ventricular end-systolic volume (LVESV) according to *NR3C2* minor (C) allele carriage. Box plots show median (line), interquartile range (box), and extreme values (whiskers).

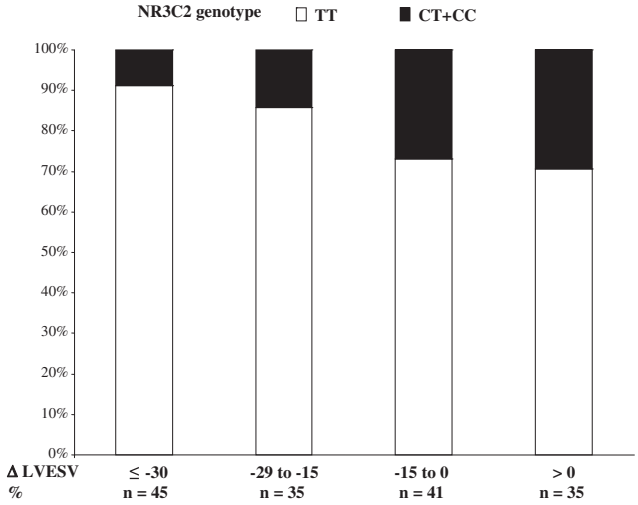

Fig. 3. Frequency percentage of *NR3C2* minor (C) allele carriage (black) according to different degrees of reverse remodeling as defined by changes in left ventricular end-systolic volume (ΔLVESV) at follow-up compared with baseline.

Table 5. Variables Associated with Lack of Reverse Remodeling by Multivariable Logistic Regression

	P Value	Adjusted Odds Ratio	95% Confidence Interval
Age	.488	0.987	0.952−1.024
Male vs female sex	.120	2.397	0.797−7.203
Previous myocardial infarction	.873	1.061	0.512−2.201
Diabetes	.055	2.579	0.981−6.781
Atrial fibrillation	.317	1.641	0.622−4.331
NYHA class III−IV vs II	.482	0.765	0.363−1.614
Left ventricular ejection fraction	.858	1.006	0.945−1.070
NR3C2 minor (C) allele carriage (CC-CT vs TT genotypes)	.014	3.093	1.253−7.632

Abbreviations as in Tables 1 and 2.

We did not assess plasma aldosterone levels or urinary sodium excretion. However, the functional effects of the polymorphism on plasma aldosterone levels may be particularly difficult to elucidate in HF patients on optimal drug therapy. In severe HF, plasma aldosterone concentrations are elevated and correlate with resistance to loop diuretics. Even under effective pharmacologic RAAS blockade with ACE inhibitors, aldosterone breakthrough occurs in a substantial proportion of HF patients with return to pretreatment plasma hormone levels[32] so large as to overcome possible differences in gene products due to polymorphisms. On the other hand, it is conceivable that biomarkers may exist that differentially interact with genotypes on phenotype expression.

Clinical Implications

The genetic findings of this pilot investigation are directly tied to the biology underlying the remodeling process. Candidate genes of the RAAS have multiple lines of evidence to support a possible role in a lack of RR: the *NR3C2* polymorphism is involved in aldosterone signaling in the myocardium and vascular wall, and blockade of the MR prevents progression of cardiac damage in the experimental model and in human disease. Nonetheless, our findings remain largely speculative unless they are proved in larger series. In particular, the role of pharmacogenomic profiling to tailor levels of aldosterone blockade requires careful investigation in adequately powered samples to account for different confounders and dosing. For this purpose, well designed registries[33] or biobanks from randomized controlled trials may prove to be more suitable.

Conclusion

Lack of RR after CRT was linked in the present series to genetic variants involved in aldosterone signaling that have been previously associated with cardiovascular phenotypes. Whether genetic characterization might better identify HF patients who may be most likely to benefit from device implantation deserves further investigation.

Acknowledgments

The authors gratefully acknowledge the expert secretarial assistance of Elisabetta Spagnolo.

Disclosures

None.

References

1. Bristow MR, Saxon LA, Boehmer J, Krueger S, Kass DA, de Marco T, et al, Comparison of Medical Therapy, Pacing, and Defibrillation in Heart Failure (COMPANION) Investigators. Cardiac resynchronization therapy with or without an implantable defibrillator in advanced heart failure. N Eng J Med 2004;350:2140−50.
2. Cleland JG, Daubert JC, Erdmann E, Freemantle N, Gras D, Kappenberger L, et al, Cardiac Resynchronization-Heart Failure (CARE-HF) Study Investigators. The effect of cardiac resynchronization on morbidity and mortality in heart failure. N Engl J Med 2005; 352:1539−49.
3. St John Sutton MG, Plappert T, Abraham WT, Smith AL, DeLurgio DB, Leon AR, et al, Multicenter In-Sync Randomized Clinical Evaluation (MIRACLE) Study Group. Effect of cardiac resynchronization therapy on left ventricular size and function in chronic heart failure. Circulation 2003;107:1985−90.
4. Verhaert D, Grimm RA, Puntawangkoon C, Wolski K, De S, Wilkoff BL, Starling RC, et al. Long-term reverse remodeling with cardiac resynchronization therapy. Results of extended echocardiographic follow-up. J Am Coll Cardiol 2010;55:1788−95.
5. Ghio S, Freemantle N, Scelsi L, Serio A, Magrini G, Pasotti M, et al. Long-term left ventricular reverse remodeling with cardiac resynchronization therapy: results from the CARE-HF trial. Eur J Heart Fail 2009;11:480−8.
6. Ypenburg C, van Bommel RJ, Borleffs CJ, Bleeker GB, Boersma E, Schalij MJ, et al. Long-term prognosis after cardiac resynchronization therapy is related to the extent of left ventricular reverse remodeling at midterm follow-up. J Am Coll Cardiol 2009;53:483−90.
7. Yu CM, Bleeker GB, Fung JW, Schalij MJ, Zhang Q, van der Wall EE, et al. Left ventricular reverse remodeling but not clinical improvement predicts long-term survival after cardiac resynchronization therapy. Circulation 2005;112:1580−6.
8. Foley PW, Chalil S, Khadjooi K, Irwin N, Smith RE, Leyva F. Left ventricular reverse remodeling, long-term clinical outcome, and mode of death after cardiac resynchronization therapy. Eur J Heart Fail 2011;13:43−51.
9. van Bommel RJ, Bax JJ, Abraham WT, Chung ES, Pires LA, Tavazzi L, et al. Characteristics of heart failure patients associated with good and poor response to cardiac resynchronization therapy: a PROSPECT (Predictors of Response to CRT) sub-analysis. Eur Heart J 2009;30:2470−7.
10. Vidal B, Delgado V, Mont L, Poyatos S, Silva E, Angeles Castel M, et al. Decreased likelihood of response to cardiac resynchronization in patients with severe heart failure. Eur J Heart Fail 2010;12:283−7.
11. Wikstrom G, Blomström-Lundqvist C, Andren B, Lönnerholm S, Blomström P, Freemantle N, et al, CARE-HF Study Investigators. The effects of etiology on outcome in patients treated with cardiac resynchronization therapy in the CARE-HF trial. Eur Heart J 2009;30: 782−8.
12. Gasparini M, Auricchio A, Regoli F, Fantoni C, Kawabata M, Galimberti P, et al. Four-year efficacy of cardiac resynchronization therapy on exercise tolerance and disease progression. The importance of performing atrioventricular junction ablation in patients with atrial fibrillation. J Am Coll Cardiol 2006;48:734−43.

13. Arshad A, Moss AJ, Foster E, Padeletti L, Barsheshet A, Goldenberg I, et al, MADIT-CRT Executive Committee. Cardiac resynchronization therapy is more effective in women than in men: the MADIT-CRT (Multicenter Automatic Defibrillator Implantation Trial with Cardiac Resynchronization Therapy) trial. J Am Coll Cardiol 2011;57: 813−20.
14. Gasparini M, F, Ceriotti C, Galimberti P, Bragato R, de Vita S, Pini D, et al. Remission of left ventricular systolic dysfunction and of heart failure symptoms after cardiac resynchronization therapy: temporal pattern and clinical predictors. Am Heart J 2008;155:507−14.
15. Linde C, Abraham WT, Gold MR, Daubert C, REVERSE Study Group. Cardiac resynchronization therapy in asymptomatic or mildly symptomatic heart failure patients in relation to etiology: results from the REVERSE (Resynchronization Reverses Remodeling in Systolic Left Ventricular Dysfunction) study. J Am Coll Cardiol 2010;56: 1826−31.
16. Bleumink GS, Schut AF, Sturkenboom MC, Deckers JW, van Duijn CM, Stricker BH. Genetic polymorphisms and heart failure. Genet Med 2004;6:465−74.
17. MacRae CA. The genetics of congestive heart failure. Heart Fail Clin 2010;6:223−30.
18. Dickstein K, Vardas PE, Auricchio A, Daubert JC, Linde C, McMurray J, et al. 2010 Focused update of ESC guidelines on device therapy in heart failure. Eur Heart J 2010;31:2677−87.
19. Zhu X, Bouzekri N, Southam L, Cooper RS, Adeyemo A, McKenzie CA, et al. Linkage and association analysis of angiotensin I−converting enzyme (ACE)−gene polymorphisms with ACE concentration and blood pressure. Am J Hum Genet 2001;68:1139−48.
20. Benjamini Y, Yekutieli D. The control of the false discovery rate in multiple testing under dependency. Ann Stat 2001;29:1165−88.
21. Auger D, van Bommel RJ, Bertini M, Delgado V, Ng AC, Ewe SH, et al. Prevalence and characteristics of patients with clinical improvement but not significant left ventricular reverse remodeling after cardiac resynchronization therapy. Am Heart J 2010;160:737−43.
22. Fornwalt BK, Sprague WW, BeDell P, Suever JD, Gerritse B, Merlino JD, et al. Agreement is poor among current criteria used to define response to cardiac resynchronization therapy. Circulation 2010;121:1985−91.
23. Kass DA. Pathobiology of cardiac dyssynchrony and resynchronization. Heart Rhythm 2009;6:1660−5.
24. Martin DT, McNitt S, Nesto RW, Rutter MK, Moss AJ. Cardiac resynchronization therapy reduces the risk of cardiac events in patients with diabetes enrolled in the MADIT-CRT Trial. Circ Heart Fail 2011;4:332−8.
25. Fantoni C, Regoli F, Ghanem A, Raffa S, Klersy C, Sorgente A, et al. Long-term outcome in diabetic heart failure patients treated with cardiac resynchronization therapy. Eur J Heart Fail 2008;10:298−307.
26. Connell JM, MacKenzie SM, Freel EM, Fraser R, Davies E. A lifetime of aldosterone excess: long-term consequences of altered regulation of aldosterone production for cardiovascular function. Endocr Rev 2008; 29:133−54.
27. Funder JW. Aldosterone and mineralocorticoid receptors in the cardiovascular system. Prog Cardiovasc Dis 2010;52:393−400.
28. Messaoudi S, Azibani F, Delcayre C, Jaisser F. Aldosterone, mineralocorticoid receptor, and heart failure. Mol Cell Endocrinol 2012;350: 266−72.
29. Galuppo P, Bauersachs J. Mineralocorticoid receptor activation in myocardial infarction and failure: recent advances. Eur J Clin Invest. Epub ahead of print 2012 March 28. http://dx.doi.org/10.1111/j.1365-2362.2012.02676.x.
30. Fraccarollo D, Berger S, Galuppo P, Kneitz S, Hein L, Schütz G, et al. Deletion of cardiomyocyte mineralocorticoid receptor ameliorates adverse remodeling after myocardial infarction. Circulation 2011;123:400−8.
31. Kathiresan S, Larson MG, Benjamin EJ, Corey D, Murabito JM, Fox CS, et al. Clinical and genetic correlates of serum aldosterone in the community: the Framingham Heart Study. Am J Hypertens 2005;18:657−65.
32. Bomback AS, Klemmer PJ. The incidence and implications of aldosterone breakthrough. Nat Clin Pract Nephrol 2007;3:486−92.
33. Koontz JI, Haithcock D, Cumbea V, Waldron A, Stricker K, Hughes A, et al. Rationale and design of the Duke Electrophysiology Genetic and Genomic Studies (EPGEN) biorepository. Am Heart J 2009;158: 719−25.