review

Genetic polymorphisms and heart failure

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Heart failure is a complex clinical syndrome. There is evidence for a genetic contribution to the pathophysiology of heart failure. Considering the fundamental role of neurohormonal factors in the pathophysiology and progression of cardiac dysfunction and hypertrophy, variants of genes involved in this system are logical candidate genes in heart failure. In this report, genetic polymorphisms of the major neurohormonal systems in heart failure will be discussed. Studies on polymorphisms of the renin-angiotensin-aldosterone system (RAAS), adrenergic receptor polymorphisms, endothelin (receptor) polymorphisms, and a group of miscellaneous polymorphisms that may be involved in the development or phenotypic expression of heart failure will be reviewed. Research on left ventricular hypertrophy is also included. The majority of genetic association studies focused on the ACE I/D polymorphism. Initial genetic associations have often been difficult to replicate, mainly due to problems in study design and lack of power. Promising results have been obtained with genetic polymorphisms of the RAAS and sympathetic system. Considering the evidence so far, a modifying role for these polymorphisms seems more likely than a role of these variants as susceptibility genes. Besides the need for larger studies to examine the effects of single nucleotide polymorphisms and haplotypes, future studies also need to focus on the complexity of these systems and study gene-gene interactions and gene-environment interactions. ***Genet Med* 2004:6(6):465–474.**

Heart failure is a complex clinical syndrome with high morbidity and mortality.[1,2] Impairment of cardiac function activates compensatory neurohormonal mechanisms, which at a later stage may accelerate progression of heart failure.[3] Coronary heart disease and hypertension are major underlying causes of heart failure. Other frequent underlying conditions include valvular heart disease and idiopathic dilated cardiomyopathy. It is difficult to predict who will develop heart failure in response to myocardial injury. Racial differences in occurrence and outcome of heart failure[4,5] and heritability estimates of variability in left ventricular mass of 28% to 65%[6,7] suggest a genetic contribution to the pathophysiology of left ventricular remodeling and heart failure.

Many studies have been published on the role of single gene mutations in (familial) cardiomyopathies.[8,9] These Mendelian traits are by nature rare, and although important at an individual level and for the understanding of disease mechanisms, are of limited significance in terms of prediction of heart failure occurrence in the population.[10] Moreover, in patients with identical gene mutations clinical manifestations of cardiomyopathy may differ. This suggests that probably also environmental and/or other genetic factors play a role.

In this report, genetic polymorphisms of major neurohormonal systems involved in the pathophysiology of heart failure will be discussed, including the renin-angiotensin-aldosterone system (RAAS), adrenergic receptor polymorphisms, endothelin (receptor) polymorphisms, and a group of miscellaneous polymorphisms. Research on left ventricular hypertrophy is also included, because this condition often precedes heart failure. Studies on the association between heart failure and human leukocyte antigen complex alleles and pharmacogenetic studies are outside the scope of this review.

METHODS

Literature for this report was systematically identified by searching PubMed for all English-language articles published up to July 2003 related to heart failure and genetic polymorphisms. Bibliographies in articles provided further references. A two-step approach was used. First, genetic polymorphisms were identified in a search with the keywords "heart failure" and "polymorphism." Second, polymorphisms that were identified with this search, were used as keywords with the addition of one of the following keywords "heart failure," "left ventricular hypertrophy," "left ventricular mass," and "cardiomyopathy." We focused on genetic association studies.

Heart failure pathophysiology

Left ventricular dysfunction begins with an injury to the myocardium, e.g., myocardial infarction, which results in loss

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of functional cells. In response, a complex cascade of interacting hemodynamic and neurohormonal mechanisms is activated to preserve cardiac function.[3] These factors alter the shape and function of the ventricle through a process called left ventricular remodeling, in which fibrosis and myocyte damage appear to be decisive morphological alterations.[11,12] The most important regulators of this process are components of the RAAS, growth factors, and endocrine hormones such as norepinephrine.

A decreased capacity of the left ventricle to empty during systole increases diastolic wall tension. The ventricle responds by enhanced contraction, following the Frank-Starling curve.[13] Additionally, the sympathetic nervous system is activated, which provides inotropic support and maintains cardiac output.[3] Both compensatory mechanisms also lead to increased internal wall stress during diastole. In response, synthesis of myofibrillar proteins is stimulated, resulting in increased wall thickness and a reduction in ventricular wall stress and dilatation.[13] Sustained sympathetic stimulation, however, activates the RAAS and other neurohormones. The main effector of the RAAS is angiotensin II, a potent vasoconstrictor and stimulator of renal sodium reabsorption and aldosterone and vasopressin release.[14] Hypoxia, shear stress, and vasoactive hormones also stimulate the generation of endothelin, the most potent endogenous vasoconstrictor.[15] Endothelin has positive inotropic and chronotropic effects, influences salt and water homeostasis, stimulates the RAAS and sympathetic system and appears to play a role in cardiac remodeling.[16] With worsening fibrosis and cardiac myocyte degeneration, left ventricular end-diastolic pressure increases and later ejection fraction decreases.[12]

To counterbalance the pressor and volume expanding effects of vasoactive neurohormones, natriuretic peptides are generated, which exert diuretic, natriuretic, and vasodilator properties.[17] Also, the kallikrein-kinin system forms bradykinin, which results in natriuresis and vasodilatation, and stimulates the production of prostaglandins.[3] In this way, a delicate hemodynamic balance is achieved, which restores cardiac function temporarily. Long-term activation of these mechanisms, however, results in progressive deterioration of ventricular function.

Genetic polymorphisms

In common multifactorial diseases, such as heart failure, the candidate gene approach is widely used to study genetic polymorphisms.[18] This allows for the identification of gene defects directly involved in the pathophysiology of heart failure (susceptibility genes), or variants involved in modification of its phenotypic expression (modifier genes). The case-control study is the most frequently used design. Candidate genes are selected based upon their biological plausibility. Polymorphisms located in coding or promoter regions of a gene may alter the function or expression of proteins encoded by the gene. Even if these polymorphisms are not functional, they are more likely to be in linkage disequilibrium with causative alleles.[10] Polymorphisms in intronic enhancer regions, both *cis* and *trans*, can also affect expression.

Considering the fundamental role of neurohormonal factors in the pathophysiology and progression of cardiac dysfunction and hypertrophy, variants of neurohormonal genes are logical candidate genes in heart failure. Successively, we will describe the potential role of these variants as susceptibility genes and as modifier genes in heart failure due to ischemic and/or dilated cardiomyopathy and in hypertrophic cardiomyopathy. Also, studies on left ventricular dimensions will be discussed. Table 1 shows genetic polymorphisms that have been associated with one or more of these disease manifestations in at least one study.

Renin-angiotensin-aldosterone system

The RAAS is one of the major systems involved in the pathophysiology of heart failure. Because it is a complex system, effects of polymorphisms influencing the expression of one of its components may be counterbalanced by compensatory changes in other components. Therefore, for individual mutations, associations are probably modest.

Angiotensin-converting enzyme I/D polymorphism

ACE enzymatically transforms angiotensin I to angiotensin II. The human ACE gene is located on chromosome 17q23. A genetic polymorphism in intron 16 of this gene is strongly associated with serum levels of ACE.[19] It is characterized by an insertion (I) or a deletion (D) of a 287 noncoding base pair (bp) Alu repeat sequence. Its functional role has been debated. Probably, this polymorphism is in strong linkage disequilibrium with another functional mutation within the gene.[20]

Raynolds and colleagues[21] published an association between the ACE I/D polymorphism and heart failure susceptibility in Caucasians. They observed an excess of DD genotype both in subjects with ischemic and idiopathic dilated cardiomyopathy compared to organ donors. However, because their control group was not in Hardy-Weinberg equilibrium, results may have been biased. Subsequently, many conflicting studies followed. A study in 70 Chinese patients appeared to confirm the presence of an association[22] but was also not in Hardy-Weinberg equilibrium. Most studies did not find an association between the ACE polymorphism and heart failure secondary to ischemic and/or dilated cardiomyopathy in 1506 Caucasian,[23–26] 287 Chinese,[27] 281 Japanese,[28] and 724 black South African subjects.[29,30] Hence, it seems unlikely that susceptibility to ischemic or idiopathic dilated cardiomyopathy in the general population is associated with the ACE I/D polymorphism.

In contrast, studies have reported more promising results on the potential role of the ACE polymorphism as a modifier gene in heart failure, at least in Caucasian populations. Andersson found an association between poor survival and DD genotype in 194 Swedish patients with heart failure.[31] A study in 57 white American patients demonstrated that DD genotype was associated with impaired exercise tolerance.[32] Others found that the D allele was associated with an increased risk of death or

Table 1

Genetic polymorphisms that have been associated with heart failure occurrence (susceptibility gene), phenotype (modifier gene), and/or left ventricular structure in genetic association studies

Genetic polymorphism	Risk allele[a]	Heart failure occurrence[c]	Heart failure phenotype[c]	DCM/DCM phenotype	HCM/HCM phenotype	LVH/LV dimensions[d]
Renin-angiotensin-aldosterone system						
ACE I/D	D	+	+	+	+	+
AGT M235T	T	+	−	−	+	+
AGT-G6A	G	+	−	−	−	+
AGT T174M	M	−	−	−	−	+
AT1R A1166C	C	−	+	−	+	+ (A)
AT2R A3123C	A	−	−	−	+	−
AT2R G1675A	A	−	−	−	−	+
CYP11B2 C-344T	C	−	−	+	+	+
Sympathetic system						
A2AR α_{2c} Wt/Del322–325	Del	+	−	−	−	−
B1AR Ser49Gly	Ser	−	+	+	−	−
B1AR Arg389Gly	[b]	−	+	+	−	+
B1AR T-2146C	C	−	−	+	−	−
B2AR Arg16Gly	Gly	−	+	−	−	+ (Arg)
B2AR Gln27Glu	Glu	−	−	−	−	+
B2AR Thr164Ile	Ile	−	+	−	−	+
Endothelin						
END1 G8002A	A	−	−	−	+	−
ETAR C1363T	T	−	−	+	−	−
ETAR C69T	T	−	−	+	−	−
Miscellaneous						
Bradykinin receptor[106]	+9	−	−	−	−	+
CMA A-1903G[37,42,107]	A	−	−	−	+	+
TNF G-308A[72,108,109]	A	−	−	+	+	−
TGFB1 Leu10Pro[110]	heterozy	+	−	+	−	−
SOD2 Ala16Val[111]	Val	−	−	+	−	−
PAF G994T[112,113]	T	−	−	+	+	−
CCR2 Val64Ile[114]	Ile	+	−	−	−	−
NOS3 Glu298Asp[115]	Asp	−	+	−	−	−
APOE ε2/ε3/ε4[116–118]	ε4	+	−	−	−	+
α-adducin Gly460Trp[58]	Trp	−	−	−	−	+
GNB3 C826T[119,120]	T	−	−	−	−	+
AMPD1+/−[121]	+	−	+	−	−	−

[a]In majority studies, [b]indecisive, [c]heart failure with multiple causes, not exclusive in DCM or HCM, [d]in patients without heart failure.

+, at least one positive study; −, no positive association studies; DCM, dilated cardiomyopathy; HCM, hypertrophic cardiomyopathy; LVH, left ventricular hypertrophy; ACE, angiotensin converting enzyme; AGT, angiotensinogen; AT1R, angiotensin II type 1 receptor; AT2R, angiotensin II type 2 receptor; CYP11B2, alodosterone synthase gene; α2-AR, α2-adrenergic receptor; B1AR, β1-adrenergic receptor; B2AR, β2-adrenergic receptor; END1, endothelin-1; ETAR, endothelin type A receptor; CMA, cardiac chymase; TNF, tumor necrosis factor α; TGFB1, transforming growth factor β-1; SOD, superoxide dismutase; PAF, platelet-activating factor; CCR, chemokine receptor; NOS, nitric oxide synthase; APOE, apolipoprotein E; GNB3, G-protein β3 subunit; AMPD1, adenosine monophosphate deaminase 1.

heart transplantation in 328 American patients.[33] Two small studies in 90 Czech and 84 Turkish subjects did, however, not detect an association between heart failure phenotype and ACE genotype.[24,26] Studies in Japanese[28,34] and Chinese patients[35] failed to associate this polymorphism with heart failure phenotype, whereas studies in blacks were contradictory. Candy et al.[30] found an association between DD genotype and reduced cardiac function and increased cavity size in South African patients with cardiomyopathy, whereas others did not.[29]

The first study on the ACE polymorphism and hypertrophic cardiomyopathy found an excess of DD genotype in Caucasian patients, especially in families with a history of sudden cardiac death.[36] Other studies have since then confirmed the higher frequency of DD genotype in hypertrophic cardiomyopathy in white and Japanese patients.[37–39] Yamada et al.[28] did not detect an association between ACE genotypes and hypertrophic cardiomyopathy in Japanese. In addition, no association was found between ACE genotypes and echocardiographic measurements. Also, a study in 104 Dutch patients did not support a role for the I/D polymorphism as a modifier in this disorder.[40] An association of ACE genotype with phenotypic expression of hypertrophic cardiomyopathy was, however, observed in Caucasians by others.[41] A study conducted in a family affected by a single mutation in the myosin binding protein-C (MyBP-C) gene confirmed these results.[42] It seems that the influence of ACE genotype in patients with hypertrophic cardiomyopathy depends on the specific disease-causing mutation, which may offer an explanation for the conflicting results.[43] Similar to ischemic and idiopathic dilated cardiomyopathy, it is more likely that the ACE I/D polymorphism acts as a modifier in hypertrophic cardiomyopathy than as a susceptibility gene.

Many studies have investigated the influence of the ACE polymorphism on left ventricular hypertrophy. There is considerable controversy on this topic. Schunkert and colleagues described an association between DD genotype and electrocardiographic left ventricular hypertrophy in 1428 Europeans.[44] Another large-scale investigation was performed in the Framingham Heart Study in 2439 subjects.[45] No association was found between ACE genotypes and echocardiographic left ventricular hypertrophy. Kuznetsova et al.[46] have summarized the majority of studies ($n = 28$, overall sample size 6638) in a metaanalysis. There was significant heterogeneity among studies and the definition of left ventricular hypertrophy varied widely. Using data from 12 case-control studies, the pooled odds ratio (OR) of left ventricular hypertrophy with presence of the D allele was 1.09 [95% confidence interval (CI) 0.98–1.21]. Subgroup analyses, however, showed that in untreated hypertensives, the risk of cardiac hypertrophy for the DD genotype compared to the II genotype was significantly increased (OR 2.92; 95% CI 1.50–5.70). Also, left ventricular mass as a continuous trait was only associated with ACE genotype in never-treated hypertensives. Therefore, findings suggest that the ACE polymorphism may only have an impact on cardiac dimensions in hypertensives. From a clinical perspective, one might speculate that this effect can be neutralized by the use of antihypertensive drugs. Although there are many studies on the ACE I/D polymorphism, study populations are often not in Hardy-Weinberg equilibrium. There may be various explanations, including differential mortality, but also genotyping error cannot be excluded. This calls for a different approach in which multiple markers are tested that are more robust than the original ACE I/D variant.

Angiotensinogen M235T polymorphism

Angiotensinogen is converted to angiotensin I. The human angiotensinogen gene is located on chromosome 1q42–43.[47] Three polymorphisms have been studied: two mutations in exon 2 resulting in a methionine to threonine exchange at position 235 (M235T) and a threonine to methionine substitution at position 174 (T174M), and a G-6A variant, which represents a guanine to adenine substitution 6 bp upstream from the initiation site of transcription in the promoter region.[48] The G-6A variant is in close linkage disequilibrium with the M235T polymorphism and will not be discussed separately. As the T174M variant has been studied only once with respect to cardiac dimensions,[49] we focus on the M235T polymorphism.

The 235T allele is associated with a stepwise increase in angiotensinogen levels and with a moderate increase in risk of hypertension.[50] Nevertheless, most studies did not find an association between the M235T polymorphism and development or progression of ischemic and dilated cardiomyopathy.[25,28,29,35] Therefore, it is unlikely that this polymorphism plays a role in these disorders. An increased risk of heart failure in subjects carrying both 235M/T and −6G/G genotypes has been reported, however, suggesting the presence of a disease-related haplotype.[51]

In hypertrophic cardiomyopathy, conflicting findings have been published. In Japanese subjects, one study revealed a higher frequency of the 235T allele in hypertrophic cardiomyopathy.[38] This could not be confirmed by others.[28] A study by Ortlepp and colleagues in 26 family members carrying a single mutation in the MyBP-C gene revealed a significant association between the angiotensinogen polymorphism and cardiac hypertrophy.[42] In contrast, the extent of hypertrophy in 108 unrelated Canadian patients was not influenced by angiotensinogen genotype.[52]

Most studies on left ventricular hypertrophy have been performed in Caucasians.[48,49,53–58] The majority did not find an association.[49,54–58] In contrast, studies in Chinese dialysis patients,[59] South Korean patients with chest discomfort or hypertension[60] and Japanese outpatients of a cardiovascular clinic[61] found an association between TT genotype and cardiac hypertrophy. This suggests a more important role for the M235T polymorphism in Asian populations, corresponding with a higher frequency of the T allele in these populations.[35] A study in 103 Japanese patients, however, did not show an association between this polymorphism and left ventricular remodeling after myocardial infarction.[62]

Angiotensin II receptor polymorphisms

The gene for human angiotensin II type 1 receptor (AT_1R) is located on chromosome 3. Stimulation of the AT_1R results in vasoconstriction, increased atherogenicity, inflammation, growth, proliferation, or coagulation, depending on local conditions.[63] In chronic heart failure, the AT_1R is downregulated in the heart. Various polymorphisms have been detected in this receptor, of which an adenine/cytosine (A/C) substitution located at position 1166 has been associated with heart failure and cardiac dimensions. This variant is probably not functional.[40] The C allele is considered to be the risk allele.

A potential interaction between ACE DD and AT1R AC/CC genotypes was described as a predictor of survival in Swedish patients with heart failure.[31] A large study in a French population (CARDIGENE study) failed to detect an association between the AT_1R polymorphism and idiopathic dilated cardiomyopathy.[25] This study, however, used unconditional analyses despite matching of cases and controls on gender and age-distribution. Sanderson and colleagues[35] did find an association between clinical course of heart failure and the C allele in 82 Chinese patients.

One study has been performed on hypertrophic cardiomyopathy in Japanese subjects. Although there was a significant difference in allele frequencies between patients and relatives, no difference in genotype frequencies was found between patients and healthy controls.[64] However, controls were not in Hardy-Weinberg equilibrium, which may have biased the results, and statistical analysis was performed unconditional, despite matching on age and gender. Two studies in Caucasian populations also found no association between the AT_1R polymorphism and occurrence of hypertrophic cardiomyopathy.[40,52] Some studies showed that the 1166C allele might adversely affect the phenotypic expression of hypertrophic cardiomyopathy.[40,42] Therefore, A1166C polymorphism does not seem to increase heart failure susceptibility, but may play a modifying role. As only one out of six studies found an association between this variant and cardiac hypertrophy, a role for the A1166C polymorphism in left ventricular remodeling seems unlikely, at least in Caucasians. Moreover, this positive study was small, and unexpectedly, the A allele was more frequent in cases.[53]

The angiotensin II type 2 receptor (AT_2R) gene is located on the X chromosome. The AT_2R probably counteracts the effects of AT_1R.[63] Deinum investigated the association between a polymorphism in exon 3 (A3123C) of the AT_2R gene and extent of hypertrophy in 103 Dutch patients with hypertrophic cardiomyopathy. The extent of echocardiographic hypertrophy decreased with the number of C alleles in women.[65] A potential interaction between AT_1R A1166C and AT_2R A3123C polymorphisms was detected in men only.

Erdmann and colleagues[66] found no significant differences between allele frequencies of a G1675A polymorphism in intron 1 of the AT_2R in 107 patients with hypertrophic cardiomyopathy, 95 patients with dilated cardiomyopathy, and 160 normal controls. In 120 young white males, however, an association between the 1675A allele and structural cardiac changes was found in mildly hypertensive persons.[67] Two independent studies on left ventricular hypertrophy in Glasgow residents showed inconsistent results.[68] As the G1675A polymorphism is located in a gene region that is involved in the transcriptional control of the AT_2R gene, this variant is potentially functional.[67,68] Therefore, more studies are needed to determine the role of this polymorphism in heart failure.

Aldosterone synthase C-344T polymorphism

Aldosterone excess is a well-documented cause of hypertension and there is convincing evidence that mineralocorticoids have adverse effects in heart failure.[69] The key enzyme in aldosterone synthesis is aldosterone synthase. The corresponding gene CYP11B2 is located on chromosome 8.[70] A cytosine/thymidine (C/T) substitution in the 5' promoter region at location −344 of the CYP11B2 gene has been identified. The functionality of this variant is unclear.[70] Few studies have described the association between heart failure and C-344T polymorphism. Studies in Caucasian and Japanese patients did not find an association between this variant and idiopathic dilated cardiomyopathy.[25,71] However, the C allele was associated with increased left ventricular volume in the Japanese patients.[71] This finding could not be confirmed in black South Africans with heart failure.[29] Studies in hypertrophic cardiomyopathy also yielded conflicting results.[42,72] Consequently, there is no firm evidence for a role of this polymorphism in heart failure.

An association between the C-344T variant and left ventricular hypertrophy was detected in young healthy Finns.[73] Although one small study confirmed this association,[74] other larger studies failed to detect a significant effect of this polymorphism on left ventricular structure.[75,76] Mayosi et al.[77] investigated the contribution of several markers in the CYP11B2 gene, including the C-344T polymorphism, as individual variants and haplotypes. Polymorphisms and haplotypes at the CYP11B2 locus were associated with a small but significant effect on variation in septal wall thickness and left ventricular cavity size (variants G5937C and A4450C respectively). No significant association was detected with cardiac mass.

Sympathetic system

Adrenergic receptors are divided into α- and β-adrenergic receptors. α_1-Adrenergic receptors are mediators of cardiomyocyte hypertrophy, whereas α_2-adrenergic receptors are presynaptic inhibitors of norepinephrine release.[78] β_1- and β_2-adrenergic receptors increase cardiac inotropy and chronotropy, the β_1-receptor being the dominant subtype.[79] In heart failure, chronic sympathetic activation leads to selective downregulation of β_1-adrenergic receptors and uncoupling of β_1- and β_2-adrenergic receptors, markedly blunting both signaling pathways.[78] Therefore, one may hypothesize that genetic variants of adrenergic receptors play a role in heart failure. In addition, β-adrenergic receptor blockers have been found to improve symptoms and mortality in heart failure, albeit with substantial interindividual variation.[79]

α_2-Adrenergic receptor Del322-325 polymorphism

The Del322-325 polymorphism in the gene for the α_{2c}-adrenergic receptor, located on chromosome 4 and leading to the deletion of four consecutive amino acids, causes a substantial loss of agonist-mediated receptor function in vitro.[80] So far, only one study addressed the potential role of this polymorphism in heart failure susceptibility. Small et al.[81] found that black subjects who were homozygous for the α_{2c} Del322-325 variant were more than 5 times as likely to have heart failure as those who were not. A two-locus analysis indicated a significant interaction between the α_{2c}Del322-325 polymorphism and the 389Arg variant of the β_1-adrenergic receptor. Although not significant, associations for white subjects were similar in magnitude and direction. The α_{2c}Del322-325 variant was more than 10 times more common in black than in white controls. As this polymorphism potentially changes the functionality of the α_{2c}-adrenergic receptor, it is an important candidate for further research on genetic factors in heart failure.

β_1-Adrenergic receptor polymorphisms

Two major polymorphic loci have been identified in the gene coding for the β_1-adrenergic receptor located on chromosome 10. A guanine/cytosine substitution at nucleotide 1165 produces a change in the amino acid sequence, substituting a glycine (Gly) for an arginine (Arg) residue at position 389. This residue lies within the intracellular carboxy-terminus that is critical for G-protein coupling. Data indicate that this polymorphism has functional consequences for intracellular signaling.[82] The second variant, an adenine/guanine substitution at nucleotide position 145, results in a serine (Ser) or glycine (Gly) at amino acid 49.[83] Genetic variation in this residue has been shown in vitro to affect receptor desensitization and agonist-dependent down-regulation.[84] So, both polymorphic loci may lead to functional changes of the β_1-adrenergic receptor and hence both may play a role in heart failure. Genetic association studies, however, have been contradictory.

In the CARDIGENE study, no association was found between the Arg389Gly polymorphism and idiopathic dilated cardiomyopathy and disease severity.[85] Others have confirmed the lack of an association with heart failure occurrence.[34,81,86] Small and colleagues[81] did find a significant interaction between the α_{2c}Del322-325 variant and the β_1 Arg389 allele in heart failure susceptibility. An impaired exercise performance was demonstrated in patients with ischemic or idiopathic dilated cardiomyopathy, who were homozygous for Gly389 compared to Arg389 homozygotes, with an intermediate level of performance in heterozygotes.[87] This may suggest a role for the Arg389Gly polymorphism as a disease modifier. Also, a study in patients with renal disease found higher cardiac mass in Gly389 homozygotes.[88] In contrast, a study in Japanese cardiomyopathy patients showed a protective effect of the Gly389 allele in susceptibility to ventricular tachycardia.[34]

Few studies have been published on the Ser49Gly polymorphism. A study in dilated cardiomyopathy demonstrated that 5-year transplant-free survival was worse in Ser49 homozygotes than in patients with the Ser49Gly variant.[83] Allele frequencies in patients did not differ from those in normal controls. Another study found impaired exercise capacity in patients with ischemic or dilated cardiomyopathy homozygous for the Ser49 allele compared to Gly49 carriers.[87] In contrast, Podlowski and colleagues[86] only found the Gly49 mutation in patients with idiopathic dilated cardiomyopathy and not in healthy volunteers.

One other polymorphic locus in the gene for the β_1-adrenergic receptor, T-2146C, has been associated with idiopathic dilated cardiomyopathy.[89] Thus far, there is not enough evidence to draw firm conclusions about the role of β_1-adrenergic receptor polymorphisms in heart failure.

β_2-Adrenergic receptor polymorphisms

An intronless gene on chromosome 5 encodes the β_2-adrenergic receptor. Attention has focused on three polymorphisms that display altered receptor function in experimental studies.[79,90] The most functionally altered receptor is due to a threonine (Thr) to isoleucine (Ile) switch at amino acid 164. This rare variant receptor exhibits abnormal coupling to stimulatory G-protein.[91] The other two polymorphic loci occur on amino acid positions 16 [arginine (Arg)/glycine (Gly) substitution] and 27 [glutamine (Gln)/glutamic acid (Glu) substitution].[90] There is marked linkage disequilibrium between these two polymorphisms.[92] The Gly16 receptor displays enhanced agonist-promoted downregulation, whereas the Glu27 form is resistant to downregulation.[90,93]

Two studies have investigated the effect of β_2-adrenergic polymorphisms in heart failure. Especially the Thr164Ile variant seems to be important in its phenotypic expression. The first study found no difference in frequency of the three polymorphisms between heart failure patients and normal subjects.[90] However, patients who were heterozygous for the Ile164 mutation had significantly worse survival than homozygous Thr164 patients. Trends for Arg16Gly and Gln27Glu did not reach statistical significance. The second study demonstrated that patients with heart failure carrying the Ile164 allele had a lower exercise capacity than patients homozygous for Thr164.[94] Patients homozygous for the Gly16 allele also demonstrated lower exercise capacity than Arg16 homozygotes. As the Thr164Ile variant is rare, both studies did not include homozygous Ile164 subjects. A study in normotensive twins indicated that all three polymorphisms might be associated with ventricular wall thickness.[92] However, other studies have failed to detect any association between the Arg16Gly and Gln27Glu polymorphisms and cardiac dimensions.[95–97] These studies did not investigate the role of the Thr164Ile variant.

Endothelin

There is considerable evidence to support a role for the endothelin system in heart failure.[98] The endothelin A receptor mainly mediates the vasoconstrictor effects of endothelin-1, the predominant endothelin isoform. The endothelin B receptor has similar affinity for all isoforms and mediates vasodila-

tation in endothelial cells and vasoconstriction in smooth muscle cells.[16] Few genetic association studies have been performed.

Studies on dilated cardiomyopathy did not detect a role of genetic polymorphisms in the endothelin-1 gene.[99,100] Brugada and colleagues showed that a G8002A polymorphism located in the 4th intron of the endothelin-1 gene on chromosome 6 might act as a modifier gene in hypertrophic cardiomyopathy.[52] Two variants of the endothelin A receptor have been studied with promising results. In the CARDIGENE study, a cytosine/thymidine (C/T) substitution in exon 8 at nucleotide position 1363 was associated with idiopathic dilated cardiomyopathy.[99] Individuals homozygous for the T allele were at significantly increased risk for this disease. A C/T substitution in exon 6 at position 69, which does not alter the amino acid sequence of the receptor, has been associated with survival in patients with nonischemic dilated cardiomyopathy.[100] Carriers of the T allele had a more than 5-fold increased risk of death within 2 years after the diagnosis. A study in 528 never-treated hypertensives demonstrated that variants in the genes encoding endothelin-1 and the endothelin A receptor are not significant determinants of cardiac morphometric parameters.[101] Although several studies also investigated the role of endothelin B receptor polymorphisms, no significant associations were found.[99–101]

Miscellaneous genetic polymorphisms

Multiple other polymorphisms have been investigated in heart failure, with variable success. These are mainly genetic variants of factors that play a role in cardiac remodeling, inflammation, signal transduction, or protection from oxidative damage. Genetic polymorphisms that have been associated with heart failure, heart failure phenotype, and/or left ventricular structure are presented in Table 1. However, few studies have been published for most of these variants and their exact role in heart failure susceptibility and modification needs to be elucidated further.

DISCUSSION

There is substantial evidence that genes play a role in the pathophysiology of heart failure. Numerous studies have investigated the association of heart failure with polymorphisms in candidate genes. Even more studies examined the role of genetic variants in left ventricular hypertrophy. Most studies focused on the ACE I/D polymorphism. In addition, genetic polymorphisms have been studied of other RAAS components, adrenergic receptors, endothelin-1, and endothelin receptors and of factors that play a role in cardiac remodeling, inflammation, signal transduction, or protection of cells from oxidative damage. Still, many genes in heart failure remain to be discovered. So far, genetic association studies in heart failure and cardiac remodeling have been highly inconsistent. As heart failure is a complex trait, there are probably several genetic variants that together result in the expression of its pathological phenotype.[79] Therefore, for individual polymorphisms, associations are likely to be modest. The same holds for variants associated with cardiac hypertrophy. The ultimate evidence of these genetic polymorphisms being more than just risk markers depends on the characterization of intermediate phenotypes that can be linked to the disease.[102] Because most studies on heart failure susceptibility did not find an association, a role for neurohormonal polymorphisms as modifier genes seems more likely.

Genetic associations that were found in initial studies have often been difficult to reproduce. There are several potential explanations for this. The major problem has been lack of power to detect the typically small effects in genetic association studies of multifactorial traits.[103] Besides increasing the sample size of a study population, this problem may be resolved by studying the combination of several genetic markers into haplotypes.[77] Small studies with statistically significant associations are more readily published. This may overestimate the true effect. Several studies used unconditional statistical methods for data analysis, despite the fact that they matched their cases and controls on population characteristics.[25,31,34,37,64,71,85] This may underestimate the effect of the polymorphisms studied, as it will bias the results toward the null hypothesis.

An association between a genetic polymorphism and a disease (phenotype) may merely be caused by its linkage disequilibrium with a mutation of a nearby gene that is the actual functional gene. Patterns of linkage disequilibrium can vary significantly within and between populations due to several factors, including population admixture and age of the mutation.[18] If a polymorphism is not functional, varying degrees of linkage disequilibrium may explain variations between populations. Much emphasis has been put on the role of population stratification in the generation of false-positive results. When the population under study consists of a mixture of subpopulations that have different allele frequencies and disease risks, genetic associations can be confounded by population stratification.[103] The most important confounder in this respect is ethnicity. Frequency and outcome of heart failure differ significantly between races,[4,5] as do allele frequencies of genetic polymorphisms.[28,35,38,80] Nearly all populations are confounded by genetic admixture at some level.[104] Lack of replicability of an association in different ethnic groups does not rule out the possibility of a causal association, because of potentially different background risks, allele frequencies, and environmental factors. Genotyping error may also affect the results of genetic association studies and is the most common cause of deviations from the Hardy-Weinberg equilibrium. An important example of this type of exposure misclassification is the underestimation of ACE I/D heterozygotes that may occur with the conventional genotyping method.[105] Most studies on heart failure used this non–I-allele–specific method for ACE genotyping may have underestimated the effect of the DD genotype. In addition, large heterogeneity in outcome measures between studies may account for contradictory findings.

Clinical application

This review raises the question whether a clinical geneticist's evaluation is indicated for predicting the occurrence of heart

failure in an individual. The general answer is no, as there is too little known currently about the role of susceptibility genes in heart failure, except for single gene mutations associated with the familial cardiomyopathies. Familial cardiomyopathies are usually detected through the clinical setting, an echocardiogram, and the presence of a family history showing dominant inheritance.[122,123] An extended family pedigree for a patient with heart failure not associated with a familial cardiomyopathy may be useful in the research setting but has no place in general medical care, and referral to a genetics clinic for this type of heart failure is not warranted at the present time. Furthermore, testing for the various genetic polymorphisms discussed in this review is not indicated in the routine evaluation of a patient with heart failure not associated with a cardiomyopathy. As a result, it will be difficult to provide accurate and knowledgeable genetic counseling for most heart failure patients not associated with a familial cardiomyopathy.

In summary, genetic association studies on heart failure and cardiac remodeling have focused on polymorphisms that may influence neurohormonal factors. Initial genetic associations have often been difficult to replicate, mainly due to faulty study designs and lack of power. Most promising results have been obtained with polymorphisms of RAAS and sympathetic system. A role for these polymorphisms as modifier rather than susceptibility genes seems more likely considering the evidence so far. Heart failure is probably caused by many genetic factors that are all components of larger complex systems and interact with environmental factors. Besides the need for larger studies to examine the effects of single genetic variants and haplotypes, future studies also need to focus on the complexity of these systems and study gene-gene interactions and gene-environment interactions.[100–121]

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