The Adrenergic Pathway and Heart Failure

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ABSTRACT

Heart failure represents the endpoint to many triggering cardiovascular pathologies. However, there are molecular and biochemical features that remain common to the failing heart, despite the varying etiologies. Principal among these is heightened activation of the sympathetic nervous system and associated enhancement of adrenergic signaling pathways via the catecholamines, norepinephrine and epinephrine. During heart failure, several hallmark alterations in the adrenergic system contribute to loss of cardiac function. To specifically study these changes in a physiologically relevant setting, we and others have utilized advances in genetically engineered mouse technology. This chapter will discuss the many transgenic and knockout mouse models that have been developed to study the adrenergic system in the normal and failing heart. These models include genetically manipulated alterations of adrenergic receptors, linked heterotrimeric G proteins, and the regulatory G protein-coupled receptor kinases (GRKs). Among the more-interesting information gained from these models is the finding that inhibition of a particular GRK — GRK2 or β adrenergic receptor kinase 1 (βARK1) — is a potential novel therapeutic strategy to improve function in the setting of heart failure. Furthermore, we will discuss recent transgenic research that proposes an important role for hypertension in the development of heart failure. Overall, genetically engineered mouse models pertaining to this critical myocardial signaling system have provided novel insight into heart function under normal conditions and during states of dysfunction and failure.

I. Heart Failure and Sympathetic Nervous System Signaling

More than 500,000 new cases of heart failure are reported each year in the United States alone, making it one of the world’s most-prolific diseases. The principal function of the heart is to provide enough oxygenated blood to meet the body’s metabolic demands through cardiac output. Although the heart is adaptable to many physiological conditions, various etiologies can perturb its function, leading to ventricular dysfunction and ultimately failure. The initial response of the heart to excessive stress is to enlarge morphologically to a state known as cardiac hypertrophy. Classically, cardiac hypertrophy is defined as the physiological response of the heart to an increased workload. This hypertrophy may serve as compensatory and aid in preventing progressive deterioration of cardiac function (Grossman et al., 1975; Chein, 1999). However, often, the stress will
overwhelm the system, sending the hypertrophied heart into failure. As the disease progresses, the heart dilates and thins, becoming too weak to maintain adequate cardiac output. During these hypertrophic and failing processes, there is sustained heightened activation of the renin-angiotensin system and the sympathetic nervous system in an attempt by the body to maintain cardiac output and systemic blood pressure (Esler et al., 1997). Recent evidence suggests that the signaling pathways stimulated during the hypertrophic process, if left unchecked, participate in the pathogenesis of heart failure and may be more important in this disease process than the actual stress placed on the heart (Esposito et al., 2002; Rockman et al., 2002).

The importance of sympathetic activity, via the catecholamines norepinephrine and epinephrine, in heart failure progression and mortality is well established (Cohn et al., 1984). At a cellular level, the catecholamines act upon the heart by binding to the adrenergic receptors (ARs), which are members of the superfamily of proteins known as the G protein-coupled receptors (GPCRs) (Caron and Lefkowitz, 1993). In the heart, norepinephrine principally binds to the $\alpha_{1B}$- and $\beta_1$AR, while epinephrine is a ligand for both $\beta_1$- and $\beta_2$AR (Caron and Lefkowitz, 1993). The $\beta_1$AR is the most-abundant $\beta$AR in the human heart, approaching 75% of the total number of receptors (Brodde, 1993). The $\beta$ARs are coupled primarily to the heterotrimeric G protein, Gs, to stimulate adenylyl cyclase activity. This association generates intracellular cyclic adenosine monophosphate (cAMP) and protein kinase A (PKA) activation, which regulate cardiac contractility and heart rate (Bristow et al., 1989). $\beta_2$ARs also can couple to the G protein, Gi, which can inhibit adenylyl cyclase activity and stimulate novel mitogen-activated protein kinase (MAPK) pathways in the heart through G$\alpha$ and G$\beta\gamma$ subunits (Rockman et al., 2002). Alternatively, binding of norepinephrine to the $\alpha_{1B}$AR elicits phospholipase C (PLC) activity via activation of the G protein, Gq, which is the principal G protein signaling pathway implicated in the hypertrophic response of the heart (Molkentin and Dorn, 2001).

Following agonist occupation of ARs, these GPCRs become substrates for regulation via G protein-coupled receptor kinases (GRKs), which phosphorylate activated receptor (Inglese et al., 1993). This phosphorylation facilitates binding of $\beta$-arrestins, which sterically interferes with further coupling to G proteins, thus desensitizing and uncoupling the signal. The principal GRKs involved in intracellular signaling within the heart are GRK2 (or $\beta$ adrenergic receptor kinase 1, $\beta$ARK1), GRK3, and GRK5, all of which have specific GPCR selectivity in vivo in the heart (Eckhart et al., 2000). As will be detailed later, GRK activity in the heart appears to play a critical role, especially in heart failure. The generalized signaling pathways, their regulation, and outcomes in the heart are depicted in Figure 1.

Importantly, in human heart failure, chronic activation of the sympathetic nervous system has adverse implications and can accelerate cardiac pathology...
Constant stimulation of ARs by catecholamines leads to selective AR downregulation (Bristow et al., 1982, 1993; Ungerer et al., 1993). However, both the AR and the AR are markedly uncoupled from their G proteins and effector systems (Bristow et al., 1982, 1989). The latter appears to be due to increased levels and GRK activity of ARK1 (Ungerer et al., 1993, 1994). In addition, Gi is significantly upregulated, to dampen adenylyl cyclase activation (Feldman et al., 1988). Overall, these molecular adrenergic changes in the failing human heart (summarized in Table I) lead to a marked attenuation of cardiac AR signaling.

Interestingly, genetic polymorphisms in ARs have been identified and may influence individual characteristics of heart failure (Green et al., 1993; Podlowski et al., 2000; Small et al., 2002). For example, individuals with an Ile164 allele for the AR have significantly reduced survival (Green et al., 1993) and lower exercise capacity (Wagoner et al., 2000). In addition, polymorphisms within the AR, combined with a deletion mutant found within the human α2c-AR gene, can act synergistically to increase the risk of heart failure in the black population (Small et al., 2002). These genetic studies, as well as recent heart failure drug...
trials, have indicated that there is still much to learn about alterations in the βAR system during cardiac failure. This includes recent studies demonstrating significant improvement in survival with βAR antagonist therapy in people with moderate and severe heart failure (Packer et al., 1996, 2001). This is contraindicative to the short-term effects of β blockers (Epstein and Braunwald, 1966).

The recent use of genetically engineered mice has provided unique experimental models for the study of cardiac adrenergic signaling alterations and the function of the normal and failing heart.

II. Mouse Models to Study the Cardiac Adrenergic System

A. LOSS AND GAIN OF AR EXPRESSION

1. The β1AR

Gene-targeted knockout mice with disruption and ablation of the β1AR gene generally are embryonically lethal. These mice recently have been reviewed in detail (Rohrer, 1998). Although surviving β1AR knockout mice have normal heart rates, their response to exercise is abrogated (Rohrer et al., 1996). Interestingly, despite the presence of β2ARs, there was no response to β-agonist stimulation, suggesting that β1AR is responsible for catecholamine-induced alterations in heart rate in the mouse (Rohrer et al., 1996).

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**TABLE I**

<table>
<thead>
<tr>
<th>Molecule</th>
<th>Change</th>
<th>References</th>
</tr>
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<tbody>
<tr>
<td>β1ARs</td>
<td>↓, uncoupled</td>
<td>Bristow et al., 1982, 1993;</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Ungerer et al., 1993, 1994</td>
</tr>
<tr>
<td>β2ARs</td>
<td>NC, uncoupled</td>
<td>Bristow et al., 1993; Ungerer et</td>
</tr>
<tr>
<td></td>
<td></td>
<td>al., 1993, 1994</td>
</tr>
<tr>
<td>βARK1</td>
<td>↑ mRNA + activity</td>
<td>Ungerer et al., 1993</td>
</tr>
<tr>
<td>GRK3</td>
<td>NC</td>
<td>Ungerer et al., 1994</td>
</tr>
<tr>
<td>GRK5</td>
<td>Not studied</td>
<td>—</td>
</tr>
<tr>
<td>β Arrestin 1 and 2</td>
<td>NC</td>
<td>Ungerer et al., 1993</td>
</tr>
<tr>
<td>Gαi</td>
<td>↑</td>
<td>Feldman et al., 1988</td>
</tr>
<tr>
<td>Gαs</td>
<td>NC</td>
<td>Feldman et al., 1988</td>
</tr>
</tbody>
</table>

[Abbreviations: AR, adrenergic receptor; GRK, G protein-coupled receptor kinase; NC, no change.]
Using the α myosin heavy chain (αMHC) promoter to target gene expression to adult ventricular myocardium, mice that overexpress the β₁AR in the heart have been generated (Englhardt et al., 1999). These mice, with 5- to 15-fold overexpression compared to endogenous βAR levels, exhibit a pathology that is consistent with chronic sympathetic stimulation, with a phenotype of dilated cardiomyopathy and heart failure (Englhardt et al., 1999). As will be detailed, this is in striking contrast to phenotypes observed with myocardial β₂AR overexpression.

2. The β₂AR

In contrast to the β₁AR knockout mice, gene disruption of the β₂AR does not appear to significantly alter cardiac physiology (Rohrer, 1998). This suggests that, under normal conditions, the β₂AR plays no major role in murine cardiac physiology. However, the β₂AR can significantly alter cardiac physiology when overexpressed. Transgenic αMHC-β₂AR mice generated by the Lefkowitz laboratory had greater than 200-fold overexpression of endogenous βARs. These mice possessed a biochemical and physiological phenotype that mimicked maximal βAR myocardial signaling and function (Milano et al., 1994b). Surprisingly, even though these mice have enhanced heart rates and contractility from a young age, there is minimal pathology present, even in mice greater than 1 year of age (Koch et al., 2000). Furthermore, other transgenic mice with lower levels of overexpression (i.e., 30- to 50-fold) have similar characteristics (Turki et al., 1996). However, Liggett and colleagues (2000) have reported that a “transgene dose-response” for the β₂AR often can have delayed deleterious consequences, similar to what is observed with minimal β₁AR overexpression.

There appears to be significant Gi coupling in αMHC-β₂AR mice, as demonstrated by studies utilizing the Gi inhibitory pertussis toxin (Xiao et al., 1999). These data contribute to recent findings demonstrating that signaling via β₁AR and β₂AR in the heart is fundamentally different (Rockman et al., 2002). The overall positive effects seen with transgenic β₂AR overexpression suggest that the use of genetic engineering to replace lost βARs with the β₂AR in the failing heart may be therapeutic (Maurice et al., 1999). Consistent with this, we have found that cardiac β₂AR overexpression can “rescue” a mouse model of decompensated hypertrophy and heart failure due to cardiac Gαq overexpression (Dorn et al., 1999). However, αMHC-β₂AR mice were unable to rescue other mouse models of cardiomyopathy (Rockman et al., 1998b; Freeman et al., 2001). Moreover, β₂AR overexpression leads to functional deterioration of the heart, following induction of pressure overload (Du et al., 2000).
3. The \( \alpha_{1B} \)AR

Two lines of mice have been generated using the \( \alpha \)MHC promoter and the \( \alpha_{1B} \)AR. These mice express either the wild-type receptor or a constitutively active mutant of the \( \alpha_{1B} \)AR (CAM\( \alpha_{1B} \)AR) (Akhter et al., 1997; Milano et al., 1994a). The CAM\( \alpha_{1B} \)AR mice exhibit significant myocardial hypertrophy, suggesting that \( \alpha_{1B} \)AR activation can induce cardiac changes independent of hemodynamic influences (Milano et al., 1994a). In contrast, mice overexpressing the wild-type \( \alpha_{1B} \)AR do not develop an increase in heart size, despite displaying some biochemical characteristics of hypertrophy (Akhter et al., 1997). These mice, however, show a reduced tolerance to chronic \( \alpha_{1B} \)AR stimulation, indicating that they are primed for a hypertrophic response (Iaccarino et al., 2001). Interestingly, the \( \alpha \)MHC-\( \alpha_{1B} \)AR transgenic mice have a decreased response to \( \beta \)AR stimulation (Akhter et al., 1997), which appears to be mediated via an observed increase in \( \beta \)ARK1 expression and activity (Akhter et al., 1997; Iaccarino et al., 2001) and an activation of the sympathetic nervous system (Iaccarino et al., 2001). Thus, this mouse model has led to an elucidation of molecular cross-talk between the \( \alpha_{1B} \)AR and the \( \beta \)AR systems in the heart. Finally, transgenic mice expressing the \( \alpha_{1B} \)AR under the control of its isogenic promoter exhibit myocardial hypertrophy and have a surprising loss of sympathetic activity (Zusic et al., 2001).

B. GENETIC ALTERATION OF CARDIAC G PROTEIN EXPRESSION

1. \( \gamma_{i} \)s

Transgenic mice overexpressing the stimulatory G protein \( \gamma_{i} \)s in the heart have been generated and characterized (Gaudin et al., 1995). These mice exhibit enhanced responsiveness to catecholamines and develop cardiomyopathy as they age (Geng et al., 1999), in a model reminiscent of human heart failure. Interestingly, the phenotype can be rescued by chronic administration of a \( \beta \)AR blocker (Asai et al., 1999), suggesting that this phenotype, at least partially, mimics chronic sympathetic nervous system activation and enhanced \( \beta \)AR signaling.

2. \( \gamma_{i} \)

Targeted disruption of \( \gamma_{i2} \) or \( \gamma_{i3} \) (the major Gi subtypes in myocardium) in mice revealed that there appears to be no significant role for Gi signaling in basal cardiac function or in the response to \( \beta \)AR stimulation in the normal heart (Jain et al., 2001). In contrast to these knockout results, expression of a novel Gi-coupled receptor in the heart resulted in a large decrease in myocardial force, suggesting that defects in the Gi signaling pathway may contribute to the
development of cardiac pathology (Redfern et al., 1999; Baker et al., 2001). These results are consistent with the upregulation of Gαi, contributing to human heart failure and the uncoupling of the βAR system (Feldman et al., 1988).

3. Gaq

Dorn and colleagues have described transgenic mice overexpressing Gaq in the heart (D’Angelo et al., 1997). αMHC-Gaq mice with 4-fold overexpression have cardiac hypertrophy and alterations in all of its molecular markers (D’Angelo et al., 1997). These animals, like the α1B-AR overexpressors, display abrogated αAR function. Crossbreeding the Gaq mice with transgenic mice that had 200-fold overexpression of the β2-AR worsened the Gaq phenotype (Dorn et al., 1999). However, a line of β1-AR mice with only 30-fold overexpression of the receptor rescued the cardiac hypertrophy (Dorn et al., 1999), suggesting that selective, controlled βAR enhancement may be beneficial. At higher levels of Gaq expression in the transgenic mice, severe heart failure and early death was observed, with a component of increased myocyte apoptosis (Adams et al., 1998).

4. Gaq Inhibition

Due to the importance of Gaq signaling in the development of cardiac hypertrophy, our laboratory set out to selectively inhibit this pathway in the heart. To achieve this, a specific peptide inhibitor consisting of the last 54 amino acids of the Gaq (GqI) was developed and studied (Akhter et al., 1998). This GqI peptide targets the receptor-Gaq interface, competitively inhibiting Gaq activation while not affecting Gs or Gi signaling (Akhter et al., 1998). Transgenic mice expressing the GqI peptide in the heart were shown to have attenuated responses to Gaq-coupled receptor stimulation (Akhter et al., 1998). When these animals were subjected to an experimental model of pressure overload cardiac hypertrophy, expression of the GqI peptide in the heart significantly inhibited development of the hypertrophic phenotype (Akhter et al., 1998). Thus, this study identified Gaq activation as the final common trigger for pressure overload hypertrophy. More recently, these mice have shown resistance to heart failure following chronic hypertrophic stimulus (Esposito et al., 2002), suggesting that Gaq-class specific inhibition is a novel strategy to prevent ventricular dysfunction in conditions of chronic hypertrophic stress.

C. MANIPULATION OF CARDIAC GRK EXPRESSION

1. βARK1

The importance of βARK1 (GRK2) in the cardiovascular system is clearly noted by the severe cardiac malformations and embryonic death observed
following βARK1 gene ablation (Jaber et al., 1996). The findings suggest a possible role for βARK1 in the normal migration and differentiation of myocardial cells during heart development. Heterozygous βARK1 knockout mice with 50% less βARK1 expression and activity in myocardium have no developmental abnormalities (Rockman et al., 1998b). Transgenic mice that overexpress βARK1 in the heart due to the use of the αMHC promoter have an attenuated response to catecholamine stimulation with desensitized βARs (Koch et al., 1995). This was a significant finding, as it represents the first demonstration that βARK1 could cause the functional uncoupling of βARs in vivo. Furthermore, these mice demonstrate that the upregulation of βARK1 seen in human heart failure may have significance and contribute to the pathogenesis of ventricular dysfunction. Contractile responses to angiotensin II also are abrogated in αMHC-βARK1 transgenic mice, suggesting that βARK1 may be important in other receptor systems in the heart (Rockman et al., 1996). Interestingly, βARK1 overexpression has no effect on cardiac α1BAR signaling, demonstrating GRK-GPCR selectivity in vivo (Eckhart et al., 2000).

2. Inhibition of βARK1

Since βARK1 activity is increased in heart failure and appears to play a role in uncoupling of βARs in the heart, we have studied the physiological consequences of βARK1 inhibition. To do this, a specific peptide inhibitor consisting of the last 194 amino acids of the βARK1 (βARKct) was developed and studied (Koch et al., 1993). The βARKct contains the G\(_{\beta\gamma}\) binding domain and competes with endogenous βARK1 for G\(_{\beta\gamma}\)-mediated membrane translocation, a process required for βARK1 activation on activated GPCRs (Koch et al., 1993). When the βARKct was expressed in the hearts of transgenic mice under the control of the αMHC promoter, cardiac physiology was altered in reciprocal fashion to that seen with βARK1 overexpression (Koch et al., 1995). The βARKct mice have enhanced cardiac function at baseline and an augmented response to catecholamines (Koch et al., 1995). Importantly, using a hybrid transgenic mouse strategy where βARK1 overexpression and βARKct expression occurred simultaneously, we have shown that the βARKct is, indeed, inhibiting cardiac βARK1 activity (Akhter et al., 1999).

The phenotypes of the βARK1 and the βARKct mice are consistent with our hypothesis that this GRK plays a critical role in cardiac function and potentially in cardiac pathologies. Interestingly, heterozygous βARK1 knockout mice also have a phenotype of enhanced cardiac function (Rockman et al., 1998b), demonstrating that lowering βARK1 expression or its activity can have profound in vivo effects on cardiac contractility. Moreover, hybrid mice that express the βARKct in the heart and are heterogeneous for βARK1 gene ablation have even-greater enhancement of cardiac function (Rockman et al., 1998b).
In addition to heart failure, enhanced βARK1 expression and activity has been shown to be indicative of several models of cardiac hypertrophy (Koch et al., 2000). Enhanced βARK1 activity in the hypertrophied heart has been shown to be responsible for the loss of βAR inotropic reserve seen in this pathological condition (Choi et al., 1997). To study the inhibition of βARK1 during hypertrophy, we used a novel transgenic mouse model. The βARKct was targeted to the heart using the cardiac ankyrin repeat protein (CARP) promoter, which turns off during adulthood. However CARP belongs to a family of fetal genes, such as atrial natriuretic factor (ANF), that can be reactivated in adult ventricular myocardium by stress. CARP-βARKct mice lose βARKct expression after 3 weeks of life and adult mice do not have enhanced contractility (Manning et al., 2000). However, following induction of pressure overload hypertrophy, expression of the βARKct is seen once again in the myocardium, resulting in improved βAR responsiveness and cardiac function (Manning et al., 2000).

3. Cardiac Transgenic Studies with GRK3 and GRK5

Following the profound effects seen with βARK1 manipulation in the hearts of transgenic mice, we studied the physiological consequences of GRK3 and GRK5 overexpression. These two GRKs are found normally in the heart but their overall role in cardiac signaling is not well understood, although GRK5 has been found to be upregulated in some animal models of heart failure (Ping et al., 1997; Vinge et al., 2001). Unlike βARK1, both GRK3 and GRK5 homozygous knockout mice are viable with no overt cardiac phenotype (Wess, 2000). Overexpression of these GRKs in the heart has, however, led to unexpected and interesting results that have uncovered novel aspects of GRK regulation in vivo in the heart. GRK3 (also known as βARK2) previously was thought to be an isozyme of βARK1, since it is highly homologous and appeared to have the same in vitro GPCR activity (Benovic et al., 1991; Freedman et al., 1995). However, when αMHC-GRK3 mice were generated and studied, there were no signaling alterations in the cardiac βAR system (Iaccarino et al., 1998a). This was the first demonstration in vivo that GRK3 was different from βARK1, with a unique GPCR specificity profile. Further investigation revealed that thrombin signaling in the heart was uncoupled in these mice, demonstrating that the thrombin receptors are in vivo substrates for GRK3 (Iaccarino et al., 1998a). The difference in these GRKs may lie in the Gβγ binding regions, which is the area between GRK3 and βARK1 that is the most divergent (Muller et al., 1997). Thus, there may be selective GPCR-mediated translocation of these GRKs. In hybrid transgenic mice with different GRKs overexpressed along with the α1BAR, it was found that GRK3 is also the primary kinase for desensitization of this AR in the heart (Eckhart et al., 2000).
GRK5, which is the second-highest expressing GRK in the heart, is not regulated by $G_{\beta\gamma}$ and thus would be expected to have different receptor substrates in the heart. However, like $\beta$ARK1 overexpressing mice, $\alpha$MHC-GRK5 transgenic mice had severely blunted $\beta$AR inotropic responses in vivo in the heart, demonstrating that this GRK also could desensitize cardiac $\beta$ARs (Rockman et al., 1996). These mice exhibited GPCR substrate selectivity, compared to $\beta$ARK1, as responses to angiotensin II were not altered, whereas this Gq-coupled receptor system was desensitized in mice overexpressing $\beta$ARK1 (Rockman et al., 1996). GRK5 also has some activity against cardiac $\alpha_{1B}$ ARs, again demonstrating a difference with $\beta$ARK1 (Eckhart et al., 2000). The overall significance of the findings that GRK5 may be altered in heart failure is not clear at this time but obviously could have important implications.

III. $\beta$ARK1 Inhibition and Rescue of Murine Models of Heart Failure

One interesting area where this research has led us is to investigate whether inhibition of $\beta$ARK1 activity could be a novel therapeutic strategy for improving function of the failing heart. Over the last few years, this has become possible to study in the mouse, as murine models of cardiomyopathy have been described, many of which have important manifestations of the human condition. These models have been the result of a specific gene deletion in the mouse or cardiac-specific overexpression of a heart failure-inducing transgene. Powerful information can be generated by cross-breeding $\alpha$MHC-$\beta$ARKct mice and various heart failure models to test the hypothesis that $G_{\beta\gamma}$$\beta$ARK1 inhibition could be beneficial. Simply studying the cardiac phenotype of these novel hybrid mice could give an answer and provide information on the role of $\beta$ARK1 and GRK activity in the pathogenesis of the various heart failure etiologies, specific for the different models. We have studied six different heart failure models with $\beta$ARKct mice and, for the most part, have seen overwhelming rescue. Table II summarizes our findings over the last 3–4 years using this novel genetic approach; some of the more-important findings are detailed in the next paragraph.

The first murine heart failure model rescued by any genetic manipulation was done by us with $\beta$ARKct animals (Rockman et al., 1998a). The model of heart failure was due to the gene knockout of the muscle LIM protein (MLP), a cytoarchitectural protein and conserved regulator of myogenic differentiation (Rockman et al., 1998a). The improvements made by the $\beta$ARKct in this model of dilated cardiomyopathy ($MLP^{-/-}$) included restoring cardiac chamber dilation, increasing basal contractility, and enhancing $\beta$AR function (Table II). A second model of heart failure with different characteristics of disease and rescue was due to cardiac-targeted overexpression of calsequestrin (CSQ), a high-capacity calcium-binding protein (Harding et al., 2001). These mice have much more-severe disease than the $MLP^{-/-}$ animals and experience early mortality, as
all mice are dead within 20 weeks of life. Thus, in this study, we were able to carry out a survival test. \( \beta \)ARKct expression nearly doubled the life-span of these heart-failure mice (Harding et al., 2001). In addition to survival, the \( \beta \)ARKct prevented excessive ventricular deterioration and improved cardiac function (Harding et al., 2001). Furthermore, combination of \( \beta \)-blocker therapy (metoprolol) and \( \beta \)ARKct expression was synergistic in improving survival (Harding et al., 2001). These data are particularly interesting, given the clinical promise of \( \beta \)-blocker therapy in human heart failure (Packer et al., 1996, 2001). Studies from our laboratory have also shown that chronic \( \beta \)AR blockade by carvedilol decreases the expression of \( \beta \)ARK1 in the heart and reduces cardiac GRK activity (Iaccarino et al., 1998b).

As detailed in Table II, cardiac \( \beta \)ARKct expression also has rescued other models of heart failure, including one with cardiac-targeted overexpression of a mutant form of the \( \alpha \)MHC gene (HCM) that is associated with human hypertrophic cardiomyopathy (Freeman et al., 2001). Interestingly, expression of the \( \beta \)ARKct in the Gq mice had no effect on the Gq phenotype, unlike the \( \beta_3 \)AR (Dorn et al., 1999). In the Gq model of decompensated cardiac hypertrophy,
βARK1 is not upregulated, suggesting that the βARKct is acting specifically to inhibit GRKs. However, the exact mechanism of the βARKct may involve sequestration of G_{βγ} from other signaling pathways, such as those involved in the activation of phosphoinositide-3 kinase (PI3K) (Naga Prasad et al., 2000, 2001) and I_{K,AC} channels (Clapham and Neer, 1997). The contribution of these other potential G_{βγ} effects to the salutary effects of βARKct in heart failure remains to be determined. Finally, the therapeutic benefit of the βARKct may involve enhanced signaling through other GPCRs such as angiotensin II receptors.

IV. Hypertension, the Adrenergic Pathway, and Heart Failure

The American Heart Association suggests that the presence of hypertension or high blood pressure in a patient doubles that person’s risk for developing heart failure. In essential hypertension, elevated blood pressure has been associated with increased sympathetic output (Mark, 1990), suggesting that the catecholamines and associated adrenergic pathways may be involved in this pathology. It also implicates hypertension as a potential primary component in the development of heart failure. Indeed, studies have shown that agents that reduce blood pressure, no matter what the mechanism, all appear to eventually reverse hypertrophy (Testa et al., 1996; Anker et al., 1997).

We previously discussed the importance of Gq signaling in cardiac hypertrophy and heart failure. As was detailed, transgenic mice that express the GqI peptide inhibitor of Gq can prevent the development of hypertrophy and heart failure in a pressure-overload model (Akhter et al., 1998; Espositio et al., 2002). To study the potential contribution of the vascular system and its associated alterations in blood pressure to this hypertrophic response, we developed a line of transgenic mice that express GqI in vascular smooth muscle cells under the control of the SM22α promoter (Keys et al., 2002). Following chronic Gq agonist administration, we observed an attenuation of mean arterial blood pressure and an inhibition of cardiac hypertrophy in the transgenic mice with vascular GqI expression (Keys et al., 2002). In contrast — and somewhat unexpectedly — when the GqI was expressed in the heart, neither hypertension nor hypertrophy was inhibited (Keys et al., 2002). These findings suggest that, during hypertension, the vascular system is the principal determinant of cardiac hypertrophy, rather than direct stimulation of the heart itself.

Interestingly, impairment of the vascular βAR system has been shown in human and animal models of hypertension (Feldman, 1990; Brodde and Michel, 1992). More specifically, elevations in βARK1 expression have been found in lymphocytes of hypertensive patients (Gros et al., 1997, 1999). Recently, we generated transgenic mice that express βARK1 in the vascular smooth muscle, again using the SM22α promoter (Eckhart et al., 2002b). These mice display attenuated vascular βAR signaling, an increase in mean blood pressure, and
develop cardiac hypertrophy (Eckhart et al., 2002b). This again implicates the adrenergic system in hypertrophy – and, in particular, cardiovascular βARK1 activity — which also proposes a link between hypertension and heart failure.

IV. Conclusion

The development of transgenic mouse models has provided a broader understanding of the physiological impact of individual proteins during heart failure. Overall, through our efforts detailed herein and those from other laboratories around the world, there are currently at least 75 genetically altered mouse models available to study the role of particular signaling systems in the heart (Chu et al., 2002). This review has focused on the adrenergic signaling pathway under normal conditions and during heart failure. It is evident that transgenic mice have given us insight into the role of adrenergic system in the heart that otherwise would not have been possible. In the future, we hope that this knowledge may yield novel therapeutic interventions for the treatment of cardiac disease. In fact, adenoviral-mediated delivery of the βARKct and β2AR to larger animal models of heart failure has resulted in beneficial effects (White et al., 2000; Shah et al., 2001; Tevaearai et al., 2002), suggesting that gene-therapy strategies may, indeed, target these AR abnormalities in heart failure in the coming years and offer new hope to patients suffering from this disease of epidemic proportions.

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