Chapter 13
Krüppel-like Factors in the Heart

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Abstract Despite the development of numerous therapies, heart disease is a major source of morbidity, mortality, and economic burden to society worldwide. A better understanding of the molecular underpinnings that lead to heart failure are likely to facilitate the development of novel therapies. The Krüppel-like factor (KLF) family of zinc finger transcription factors play important roles in modulating cellular functions in a broad range of mammalian cell types, and accumulating evidence demonstrates important roles of these factors in cardiovascular biology. This chapter describes our current understanding of the role of the KLF gene family in cardiac biology and the potential for these factors to serve as therapeutic targets.

Introduction

Heart disease is a major cause of morbidity and mortality worldwide (Jain and Ridker 2005) and a better understanding of the molecular mechanisms underlying its pathogenesis is extremely valuable from both scientific and therapeutic standpoints. Heart failure is a condition that results from a broad array of insults that impair the pump function of the heart, including ischemia, valvular disease, hypertension, and diabetes. Accumulating evidence provides that these stressors trigger a complex series of signaling cascades that can alter gene programs in both cardiac myocytes and interstitial tissues (Braunwald 2008). Ultimately, these alterations in cell signaling and gene expression can lead to pathological remodeling of the heart, which is characterized by hypertrophic enlargement of myocytes, interstitial fibrosis, electrophysiological abnormalities, contractile dysfunction, altered calcium homeostasis, and metabolic derangements. However, the precise molecular mechanisms by which these alterations in gene expression occur remain incompletely understood.

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There has been intense interest in the cytosolic and nuclear signaling pathways that control cardiac development and remodeling (Heineke and Molkentin 2006), and a number of key transcription factors to critical regulators of these processes have been shown (Adhikari et al. 2006; Akazawa and Komuro 2003; Epstein and Parmacek 2005; Finck and Kelly 2006; Oettgen 2006; Perry and Soreq 2002; Puigserver and Spiegelman 2003). Studies from our group and others have recently demonstrated a critical role for the Krüppel-like factor (KLF) family of zinc finger transcription factors in cardiac biology (Feinberg et al. 2004; Perry and Soreq 2002; Suzuki et al. 2005; Wei et al. 2006). This chapter focuses on the emerging role of the KLF family of transcription factors in the heart with an emphasis on the pathobiology of heart failure.

Overview of KLFs in the Heart

Although there has been an explosion of studies on KLFs in a broad variety of tissues and disease states, the number of reports describing the role of KLFs in the heart are few (Haldar et al. 2007). The published reports describing a role for KLFs in the heart are as follows: (1) KLF5 in cardiac fibroblasts (Shindo et al. 2002); (2) KLF15 in postnatal cardiomyocyte (Fisch et al. 2007) and cardiac fibroblast (Wang et al. 2008) biology; (3) KLF13 in the developing vertebrate heart (Lavallee et al. 2006); and (4) a brief report describing the cardiac phenotype of KLF10 knockout mice (Rajamannan et al. 2007). There are also two of reports profiling the expression of various KLFs in cultured cardiomyocytes in response to endothelin-1 (ET-1), oxidative stress and cytokines (Clerk et al. 2006; Cullingford et al. 2008). These published findings of KLFs in cardiac biology are summarized in Table 1. The remainder of this chapter is divided into subsections organized by each KLF family member.

KLF5 in the Heart

KLF5 (also known as BTEB2/IKLF) was cloned as a novel GC box-binding protein from a human placenta cDNA library and originally identified as a positive regulator of *SMemb*, a gene induced in activated smooth muscle cells (Sogawa et al. 1993; Watanabe et al. 1999). Elegant work from the laboratory of Ryozo Nagai has delineated the importance of this factor in cardiac and vascular biology (Shindo et al. 2002). In the heart, KLF5 is expressed primarily in cardiac fibroblasts and serves as a critical effector of angiotensin II signaling in these cells. Angiotensin II stimulation induces KLF5 expression in primary cardiac fibroblasts. Moreover, the angiotensin II mediated induction of platelet-derived growth factor-A (PDGF-A) is dependent on the recruitment of KLF5 to the PDGF-A promoter.

To further understand the role of KLF5 in cardiovascular biology, KLF5 was targeted systemically in the mouse germline (Shindo et al. 2002). KLF5 homozygous-null

Table 1 Function and regulation of KLFs in the heart

Krüppel-like factor	Function/regulation/observation
KLF2	• Expressed in cultured cardiomyocytes • Induced by ET-1 and hydrogen peroxide • Downregulated by TNF-α and IL-1β
KLF3	• Expressed in cultured cardiomyocytes • Downregulated by ET-1
KLF4	• Expressed in cultured cardiomyocytes • Induced by ET-1 and hydrogen peroxide
KLF5	• Expressed in cardiac fibroblasts • Expressed in cultured cardiomyocytes • Induced by angiotensin II • Induced by ET-1 and hydrogen peroxide • Regulates expression of PDGF-A and TGF-β • Reduced hypertrophic remodeling is seen in response to ngiotensin II infusion in KLF5 haplo-insufficient mice • Interacts with retinoic acid receptor-α
KLF6	• Expressed in cultured cardiomyocytes • Induced by ET-1 and hydrogen peroxide
KLF9	• Expressed in cultured cardiomyocytes • Induced by ET-1
KLF10	• Expressed in cultured cardiomyocytes • Induced by ET-1 and hydrogen peroxide • Mice with systemic KLF10 deficiency develop spontaneous pathological hypertrophy by age 16 months in males but not females • Regulates expression of pituitary tumor transforming gene (*Pttg1*); however, functional significance of this observation is unknown
KLF11	• Expressed in cultured cardiomyocytes • Induced by ET-1
KLF13	• Expressed in developing cardiomyocytes • Can bind and activate the BNP promoter • Can transactivate multiple cardiac promoters in concert with GATA4 • Interacts with the zinc finger domain of GATA4 • Deficiency of KLF13 in *Xenopus* leads to cardiac developmental abnormalities (atrial septal defects and ventricular hypotrabeculation) • Cardiac phenotype of KLF13-deficient *Xenopus* embryos can be rescued by GATA4 overexpression
KLF15	• Expressed in cardiomyocytes and cardiac fibroblasts • Cardiac expression is low in developing heart and dramatically upregulated postnatally • Downregulated by ET-1 in cultured cardiomyocytes • Inhibitor of cardiac hypertrophy and fibrosis • Can inhibit GATA4 and MEF2 DNA binding and transcriptional activity • Mice with systemic KLF15 deficiency develop severe eccentric hypertrophy and exaggerated cardiac fibrosis with pressure overload • Inhibits TGF-β-induced CTGF expression in cardiac fibroblasts • Represses Smad3-mediated induction of the CTGF promoter in part via its ability to inhibit PCAF recruitment

BNP = brain natriuretic peptide; CTGF = connective tissue growth factor; ET-1 = endothelin-1; IL-1β = interleukin-1β; MEF2 = myocyte enhancing factor 2; PCAF = p300/CBP-associated factor; PDGF-A = platelet-derived growth factor A; TGF-β = transforming growth factor-β; TNF-α = tumor necrosis factor-α

mice die near embryonic day 8.5 (E8.5), although the precise developmental defect in these mice has not been well characterized. KLF5$^{+/-}$ mice are viable into adulthood and demonstrate resistance to angiotensin-mediated cardiac remodeling. Mice with KLF5 haplo-insufficiency showed a blunted hypertrophic response to angiotensin II infusion with reduced cardiac mass, wall thickness, cardiac fibrosis, and PDGF-A expression (Fig. 1A) (Shindo et al. 2002). Furthermore, angiotensin II mediated induction of transforming growth factor-β (TGF-β) and collagen type IV were also blunted in KLF5 haplo-insufficient hearts. Interestingly, the investigators show that KLF5 is able to interact with the retinoic acid receptor-α (RARα), suggesting that RARα activation regulates KLF5 function. Taken together, these observations indicate that KLF5 plays an important role in cardiac remodeling and expands the repertoire of angiotensin-responsive transcription factors in the cardiovascular system (Shindo et al. 2002).

KLF15 in the Heart

KLF15 is expressed in multiple tissues, including liver, white and brown adipose, kidney, heart, and skeletal muscle (Gray et al. 2002). KLF15 has been implicated as a critical regulator of adipogenesis (Mori et al. 2005) and hepatic gluconeogenesis (Gray et al. 2007). Recently, studies from our group demonstrated that KLF15 is a novel negative regulator of cardiac hypertrophy (Fisch et al. 2007) and fibrosis (Fisch et al. 2007; Wang et al. 2008).

KLF15 expression in the developing heart is minimal and is detectable only at very low levels during the early postnatal period. However, cardiac KLF15 expression is robustly induced within the first several weeks postnatally. Interestingly, this period is a time when ANF, BNP, and cyclin-A are downregulated (Fisch et al. 2007). KLF15 levels are reduced dramatically by pressure overload in murine models and in human hearts with left ventricular hypertrophy (LVH) due to valvular aortic stenosis (Fisch et al. 2007). Consistent with this observation, various pro-hypertrophic neurohormonal agonists such as phenylephrine and ET-1 also reduce KLF15 expression in cultured cardiac myocytes (Fisch et al. 2007).

Fig. 1 (continued) show exaggerated pathological remodeling in response to left ventricular (LV) pressure overload. Hearts from KLF15 knockout mice show eccentric hypertrophy (*left panels*). M-mode echocardiography shows severe LV dilation and systolic dysfunction (*middle panels*). Isolated KLF15$^{-/-}$ cardiomyocytes are enlarged compared to those of the wild-type controls (*right panels*). (Adapted from Fisch et al. 2007, with permission. © National Academy of Sciences, 2007.) **c** KLF15 knockout mice show exaggerated collagen deposition after 1 week of ascending aortic constriction. LV sections are stained by Masson's trichrome. (From Wang et al. 2008, with permission from Elsevier.) **d** KLF13 knockdown in *Xenopus* embryo causes atrial septal abnormalities and defects in ventricular trabeculation. (From Lavallee et al 2006, with permission from Macmillan Publishers.)

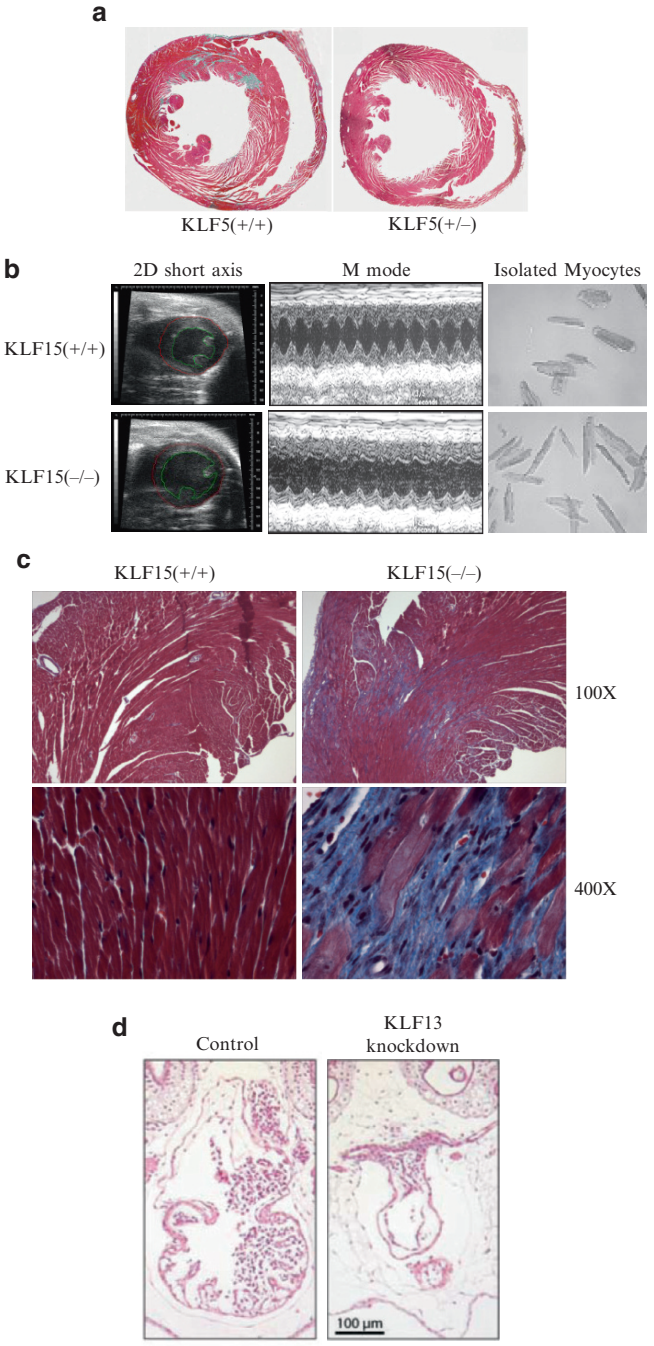

Fig. 1 Krüppel-like factors (KLFs) are important regulators of cardiac biology. **a** KLF5 haploinsufficient mice show reduced perivascular fibrosis in response to angiotensin II infusion. (From Shindo et al. 2002, with permission from Macmillan Publishers, © 2002.) **b** KLF15 knock out mice

We performed gain- and loss-of-function studies to understand the detailed role of KLF15 in the heart (Fisch et al. 2007). Overexpression of KLF15 in neonatal rat ventricular myocytes (NRVMs) inhibits the cardinal features of cardiomyocyte hypertrophy, such as cell growth, protein synthesis, and fetal gene expression. To understand the role of KLF15 in vivo, we generated systemic KLF15 knockout mice. These KLF15 homozygous null mice are viable and fertile. No overt cardiac decompensation was observed in KLF15 knockout mice at in the baseline state (age 8–12 weeks), however, these animals show exaggerated hypertrophic remodeling in response to pressure overload. Hearts from KLF15 knockout mice after aortic constriction showed cavity enlargement, impaired systolic function, and exaggerated fetal gene expression (Fig. 1B). Furthermore, cardiac myocytes from these animals were large and elongated, which is suggestive of eccentric hypertrophic remodeling (Fig. 1B).

From a mechanistic standpoint, KLF15 can attenuate transcriptional activity of MEF2 and GATA4 (Fisch et al. 2007), transcription factors that are critical hypertrophic effectors (Czubryt and Olson 2004; Pikkarainen et al. 2004), in large part via their ability to bind and transactivate promoters of key pro-hypertrophic genes. KLF15 is able to inhibit the ability of these factors to bind target promoters; and further study is underway to elucidate the precise molecular mechanism underlying the inhibitory effect of KLF15 on MEF2 and GATA4.

Interestingly, KLF15 knockout mice have cardiac phenotypes similar to those of transgenic mice overexpressing both MEF2 or GATA4 in the heart. Transgenic overexpression of MEF2A and MEF2C in the heart causes dilated cardiomyopathy in a dose-dependent manner in response to pressure overload (Xu et al. 2006). Cardiomyocytes from these MEF2-transgenic hearts have large, elongated myocytes (Xu et al. 2006), similar to the cardiomyocytes derived from KLF15 knockout mice (Fisch et al. 2007). High-level overexpression of GATA4 in the heart results in severe cardiomyopathy and premature death (Liang et al. 2001). A spontaneous cardiomyopathy (increased heart mass and hypertrophic gene expression) is observed even with modest GATA4 overexpression (Liang et al. 2001). Taken together, it is likely that these two factors are activated in the absence of KLF15 and cause exaggerated cardiac remodeling in response to stress in KLF15-deficient mice. As described before, KLF15 has an intriguing expression pattern—notably its dramatic postnatal induction. Molkentin and colleagues (Molkentin and Markham 1993) showed an increase in MEF2 binding and activity during the postnatal period. As such, the inhibitory effect of KLF15 for MEF2 raises the possibility that KLF15 plays a regulatory role in postnatal cardiac maturation (Fisch et al. 2007).

Our group has also recently identified a role for KLF15 in the cardiac fibroblast as a negative regulator of connective tissue growth factor (CTGF) signaling. CTGF is expressed in both cardiomyocytes and cardiac fibroblasts (Chen et al. 2000) and plays an important role in the development of fibrosis in disease states such as atherosclerosis (Oemar et al. 1997) and heart failure (Chen et al. 2000). TGF-β1 is a major regulator of CTGF expression (Chen et al. 2000). The TGF-β receptor is a serine/threonine kinase transmembrane heteromeric type I and type II receptor complex that signals through Smad family transcription factors upon receptor activation. Smad proteins can be divided into three groups: receptor-activated

type (Smads 1, 2, 3, 5, and 8), co-mediator type (Smads 4 and 10), and inhibitory type (Smads 6 and 7) (Khan and Sheppard, 2006). Among them, Smad3 has been shown to bind to a consensus element in the CTGF promoter by TGF-β1 stimulation (Chen et al. 2002; Grotendorst et al. 1996).

We demonstrated that KLF15 knockout mice show exaggerated collagen deposition and excess induction of CTGF (trichrome staining, Fig. 1C) in response to pressure overload (Wang et al. 2008). Furthermore, adenoviral overexpression of KLF15 inhibits CTGF induction by TGF-β1 in neonatal rat ventricular fibroblasts (NRVFs), and this repressive effect occurs at the promoter level. The electrophoretic mobility shift assay (EMSA) showed that this repressive effect was not due to inhibition of Smad3 binding to the CTGF promoter. As the protein P/CAF has been implicated as an important transcriptional co-activator of Smad3 target genes, we hypothesized that KLF15 may inhibit CTGF promoter activity via an inhibitory effect on P/CAF recruitment. Indeed, a co-immunoprecipitation assay demonstrated that KLF15 interacts with P/CAF, and a chromatin immunoprecipitation assay revealed that KLF15 overexpression inhibited recruitment of P/CAF to CTGF promoter. Moreover, repression of the CTGF promoter by KLF15 is rescued by P/CAF overexpression (Wang et al. 2008). These observations suggest that KLF15 is a negative regulator of CTGF expression in cardiac fibroblasts, in part via its ability to inhibit P/CAF–Smad3 signaling at the CTGF promoter.

KLF13 in the Heart

Expression of KLF13 (also known as FKLF-2/BTEB3) is restricted to erythroid cells, T lymphocytes, heart, and skeletal muscle (Asano et al. 2000; Song et al. 1999). KLF13 is detectable at low levels by reverse transcription-polymerase chain reaction (RT-PCR) in other adult mouse tissues (Scohy et al. 2000). Developmental expression of KLF13 is seen in the heart, cephalic mesenchyme, dermis, and epithelial layers of the gut and urinary bladder in the mouse embryo (Martin et al. 2001). Previous studies demonstrated that KLF13 plays an important role in regulation of erythroid gene expression (Feng and Kan 2005) and plays critical role in RANTES induction in activated T lymphocytes (Ahn et al. 2007; Song et al. 1999).

Nemer and colleagues demonstrated a role for KLF13 in the embryonic myocardium in studies of BNP gene regulation and *Xenopus* development (Lavallee et al. 2006). The investigators previously reported a proximal BNP promoter that can induce cardiac transcription maximally (Grepin et al. 1994). An essential KLF consensus site (CACCC) is located nearby GATA sites of this proximal BNP promoter and was shown to be essential for promoter activity. KLF13 was able to bind the CACCC element in the proximal BNP promoter, as demonstrated by electrophoretic mobility shift assay (EMSA). The authors further demostrated that KLF13 syngerizes with GATA4 to transactivate multiple cardiac promoters (BNP, ANF, β-MHC, cardiac α-actin). In addition, KLF13 was shown to be able to interact with the N-terminal zinc finger of GATA4 (Lavallee et al. 2006).

To gain a better understanding of the role of KLF13 in heart development, the expression pattern of KLF13 in the mouse embryonic heart was studied. Cardiac expression of KLF13 was first detected at E9.5. Subsequently, expression of KLF13 was seen in the developing atrial myocardium, ventricular trabeculae, atrioventricular (AV) cushions, and the truncus arteriosus. Postnatally, KLF13 expression was reduced in the heart and was restricted to the valves and interventricular septum. KLF13 knockdown in the *Xenopus* embryo was used to explore the role of KLF13 in heart development. KLF13-deficient embryos showed atrial septal defects and ventricular hypotrabeculation (Fig. 1D). This observation is consistent with the phenotype of humans with GATA4 mutation and mice with GATA4 deficiency (Epstein and Parmacek 2005). There was no correlation between this hypoplastic phenotype and increased apoptosis, suggesting that KLF13 may be involved in regulating cardioblast proliferation. Interestingly, GATA4 overexpression in these embryos could rescue these cardiac defects in a dose-dependent manner, suggesting that KLF13 and GATA4 are factors that can work synergistically in heart development. These findings using *Xenopus* as a model system indicate that KLF13 may be a novel candidate gene for human congenital heart disease.

However, we noted that the role of KLF13 in mammalian systems may be more complex. KLF13 knockout mice were recently developed in the Krensky Laboratory (Zhou et al. 2007) and were found to be viable. These investigators identified defects in T-lymphocyte survival. However, the role of KLF13 in cardiac biology has not been reported to date, .

KLF10 in the Heart

Role of KLF10/TIEG1 in the heart is not well understood. KLF10 was initially reported as a TGF-β inducible early gene 1 (TIEG-1) in osteoblasts (Subramaniam et al. 1995). Spelsberg and colleagues have shown that KLF10 plays an important role in the regulation of bone mineralization (Subramaniam et al. 2005), osteoclast differentiation (Subramaniam et al. 2005), and epithelial proliferation (Subramaniam et al. 1998; Tachibana et al. 1997). KLF10 regulates Smad signaling in osteoblasts, and KLF10 deficiency leads to osteopenia (Bensamoun et al. 2006) and impaired tendon healing (Tsubone et al. 2006).

KLF10 has been shown to be expressed in the adult heart at low levels, but its distribution within the myocardium is unknown (Subramaniam et al. 1995). In addition, its expression levels in the developing heart are not known. Spelsberg and colleagues reported the cardiac phenotype of KLF10 null mice (Rajamannan et al. 2007). These investigators observed spontaneous pathological cardiac hypertrophy in male (but not female) KLF10 knockout mice at 16 months of the age. Affected mice have increased heart mass, wall thickness, fibrosis, and myocyte disarray with preservation of LV systolic function. The exact timing of onset of this phenotype is not yet known. From a mechanistic standpoint, analysis of hypertrophic KLF10 knockout hearts revealed that KLF10 may regulate the pituitary tumor transforming

gene (Pttg1), but the significance of this finding is unclear. There are several questions remaining questions regarding KLF10's role in the heart: (1) Is KLF10 expressed in cardiomyocytes, cardiac fibroblasts, or both? (2) Is KLF10 expression altered with mechanical or neurohormonal stress? (3) What are KLF10 target genes in the heart, and what mechanisms explain the pathology seen in KLF10-null hearts? Indeed, future studies will continue to elucidate the importance of KLF10 in cardiac remodeling.

Regulation of KLFs by Hypertrophic and Apoptotic Stimuli

Recent expression-profiling studies have reported differential regulation of KLFs in cultured cardiomyocytes in response to pharmacological stimuli and oxidative stress. Clerk and colleagues reported expression profiles of KLFs in response to endothelin (ET-1) stimulation in neonatal rat cardiomyocytes. Quantitative PCR analysis revealed that expressions of KLFs 2, 4, 5, 6, 9, and 10 are induced rapidly and transiently by ET-1, whereas expressions of KLFs 3, 11, and 15 are downregulated. As oxidative stress and cytokine stimulation are implicated in cardiac myocyte apoptosis, these investigators also examined the effects of hydrogen peroxide and inflammatory cytokines on the expression of KLFs. Hydrogen peroxide upregulated KLFs 2, 4, 5, 6, and 10 mRNA expression levels and reduced KLF15 expression in cultured cardiomyocytes (Clerk et al. 2006; Cullingford et al. 2008). In addition, KLF2 is downregulated by tumor necrosis factor-α (TNF-α) and interleukin-1β (IL-1β) (Cullingford et al. 2008). Although the physiological relevance of these findings is not yet known, these observations raise the possibility that KLFs other than KLFs 5, 10, 13, and 15 may be involved in regulating cardiac growth and the response to stress.

Future Directions

Although the accumulating evidence demonstrates that KLFs play an important role in cardiac development and remodeling, there are significant questions remaining that must be addressed. First, little is known about expression profiles of KLFs in the developing and postnatal heart. In addition, distribution of KLF expression among the multiple cellular subsets that comprise the myocardium (cardiomyocytes, fibroblasts, endothelial cells, vascular smooth muscle cells, immune cells) must be defined. The relative function of KLFs in these various cell types of the heart are of great interest and will undoubtedly be important in understanding the interplay between these tissues in heart disease. Tissue- and cell-type-specific gain- or loss-of-function approaches will be necessary to address these questions.

Recent studies have highlighted the importance of coupled cardiac angiogenesis as an adaptive feature of compensated cardiac hypertrophy (Sano et al. 2007).

A number of KLFs have already been implicated in the angiogenic response. For example, KLF2 is implicated as an antiangiogenic factor in endothelial cell biology (Bhattacharya et al. 2005). It is highly likely that this family of transcription factors has broad roles in regulating this process in the myocardium under hypertrophic and ischemic conditions.

Another area in which the KLFs are likely to play an important role is in the context of cardiac metabolism. There is certainly increasing appreciation that alterations in cardiac fatty acid and glucose utilization can affect the heart's response to stress, particularly in disease states such as diabetes or obesity. Recent studies have identified several members of the KLF family as important regulators of adipogenesis, glucose homeostasis, and energy metabolism. Among the KLFs implicated in cardiac biology, KLF5 and KLF15 have been shown to alter cellular metabolism. For example, KLF5 regulates genes involved in skeletal muscle lipid oxidation and energy coupling such as UCP and CPT1—genes that certainly affect cardiac energetics. Intriguingly, Oishi and colleagues showed that this regulation occurs in cooperation with PPARδ—a nuclear receptor that has been shown to regulate cardiac fatty acid and glucose utilization (Burkart et al. 2007; Oishi et al. 2008). KLF15 has been shown to critically regulate systemic glucose homeostasis through effects on hepatic amino acid catabolism, which certainly raises the possibility that this factor has an important role in cardiac metabolism (Gray et al. 2007). Indeed, it is exciting to postulate that cooperative interactions between KLFs and PPARs—two major transcription factor families—may critically regulate cardiac substrate utilization and consequently cardiac function.

Finally, it is of utmost importance to identify compounds that regulate KLFs or interact with KLFs in the heart. For example, KLF5's function in the heart can be modulated by RARα antagonists (Shindo et al. 2002). Furthermore, clear interplay between statins and KLF2 has been demonstrated in endothelial biology (Sen-Banerjee et al. 2005). Neurohormonal antagonists that are currently used in heart failure therapy may regulate KLFs (e.g., KLF5, KLF15) in the heart. As is the case with statins and KLF2 in the endothelium, it is possible that KLFs can mediate favorable myocardial effects of drugs such as β-blockers, angiotensin-converting enzyme (ACE) blockers, and angiotensin-II receptor (AT_1) blockers. These studies have important implications for the treatment of cardiomyopathic conditions.

Another critical issue is the delineation of overlapping and restricted roles of the multiple KLFs that are co-expressed in the heart. For example, KLF13 and KLF15 modulate GATA4 activity oppositely. KLF13 synergizes with GATA4 to activate multiple promoters (Lavallee et al. 2006), whereas KLF15 inhibits induction of these promoters by GATA4 (Fisch et al. 2007). These facts raise the possibility that KLF15 may regulate GATA4 activity in part through inhibition of KLF13's function. As has been shown in other tissues, it is likely that KLFs family members regulate the expression and function of each other in the same cell type (Funnell et al. 2007). Another example of potential interplay is between KLF5 and KLF15. Both are expressed in cardiac fibroblasts: KLF5 promotes fibrosis, and KLF15 inhibits it. It is possible that a tight balance of relative expression/activity of KLFs influences the heart's response to physiological and pathological stimuli.

Hence, it is important to identify common target genes or interacting proteins for KLFs that are co-expressed in the heart to better define their overlapping or divergent roles.

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