Biochimica et Biophysica Acta 1833 (2013) 901–908

Contents lists available at SciVerse ScienceDirect

Biochimica et Biophysica Acta

journal homepage: www.elsevier.com/locate/bbamcr

Review

A-kinase anchoring proteins: Molecular regulators of the cardiac stress response ☆

Dario Diviani *, Darko Maric, Irene Pérez López, Sabrina Cavin, Cosmo D. del Vescovo

Département de Pharmacologie et de Toxicologie, Faculté de Biologie et Médecine, Lausanne, 1005, Switzerland

ARTICLE INFO

Article history:
Received 2 July 2012
Received in revised form 26 July 2012
Accepted 29 July 2012
Available online 4 August 2012

Keywords:
A kinase anchoring protein (AKAP)
Protein kinase A
Cardiac remodeling
Signaling
Cardiomyocyte

ABSTRACT

In response to stress or injury the heart undergoes a pathological remodeling process, associated with hypertrophy, cardiomyocyte death and fibrosis, that ultimately causes cardiac dysfunction and heart failure. It has become increasingly clear that signaling events associated with these pathological cardiac remodeling events are regulated by scaffolding and anchoring proteins, which allow coordination of pathological signals in space and time. A-kinase anchoring proteins (AKAPs) constitute a family of functionally related proteins that organize multiprotein signaling complexes that tether the cAMP-dependent protein kinase (PKA) as well as other signaling enzymes to ensure integration and processing of multiple signaling pathways. This review will discuss the role of AKAPs in the cardiac response to stress. Particular emphasis will be given to the adaptative process associated with cardiac hypoxia as well as the remodeling events linked to cardiac hypertrophy and heart failure. This article is part of a Special Issue entitled: Cardiomyocyte Biology: Cardiac Pathways of Differentiation, Metabolism and Contraction.

1. Introduction

Cardiovascular disease culminating in heart failure represents a leading cause of morbidity and mortality in Western societies. Heart failure can be triggered by acute or chronic stresses placed on the myocardium such as myocardial infarction, long-term hypertension, congenital heart disease, valvular disease and chronic myocarditis [1,2]. In response to these primary stresses, different compensatory mechanisms are activated in the damaged heart including hypertrophic cardiomyocyte growth, fibroblast proliferation and extracellular matrix deposition [3,4]. This causes the ventricular walls to thicken and ventricular mass to increase. These myocardial alterations, referred to as cardiac remodeling, are initially compensatory and contribute to maintain normal cardiac output [5]. However, in the long term they can lead to increased cardiomyocyte death and extensive myocardial fibrosis which can cause severe systolic and diastolic dysfunctions leading to heart failure [6].

In western societies heart failure is a syndrome that affects about 2% of the adult population and has an annual incidence of 1% of senior citizens. Since the five-year survival rate after diagnosis is lower than 50%, many efforts have been made in recent years to unravel the molecular mechanisms underlying this pathology.

It has become increasingly clear that transduction of physiological and pathological signals in the heart is controlled at the molecular level by scaffold, anchoring, and adaptor proteins. By positioning signaling enzymes to subcellular sites where they can be accessed by activators and, in turn, interact with particular substrate [7,8], these families of proteins ensure specificity and coordination of transduction events.

A-kinase anchoring proteins (AKAPs) are a family a scaffold proteins that tether the cAMP-dependent protein kinase (PKA) as well as other signaling enzymes at focal points within the cell to ensure integration and processing of multiple signaling pathways [9,10]. All AKAPs contain a PKA anchoring motif consisting of a conserved amphipathic helix that binds a dimerization and docking domain located in the extreme N-terminus of the regulatory subunit dimer of PKA [11,12]. Additional docking motifs located on the anchoring protein allow AKAPs to recruit additional signaling enzymes including kinases, phosphatases, phosphodiesterases, adenylyl cyclases, and small molecular weight GTPase to constitute multienzyme signaling complexes [7]. Subcellular targeting of individual AKAP signaling complexes at precise subcellular locations is achieved through specialized protein or lipid binding domains located on each anchoring protein [13].

2. Anchored PKA signaling in the heart

The cAMP/PKA pathway plays a crucial role in regulating cardiac contractility under physiological conditions. In this respect, β-adrenergic mediated PKA activation has been shown to regulate the phosphorylation and activity of proteins that, directly or indirectly, control sarcomere contraction and action potential duration. In particular, phosphorylation of voltage gated L-type Ca^{2+} channels and ryanodine receptor 2 (RyR2) enhances Ca^{2+} mobilization from intracellular stores, and favors contraction [14,15], whereas phosphorylation of phospholamban (PLB), a negative regulator of the activity of the

☆ This article is part of a Special Issue entitled: Cardiomyocyte Biology: Cardiac Pathways of Differentiation, Metabolism and Contraction.

* Corresponding author at: Département de Pharmacologie et de Toxicologie, Rue du Bugnon 27, 1005 Lausanne, Switzerland. Tel.: +41 21 692 5404; fax: +41 21 692 5355.
E-mail address: Dario.diviani@unil.ch (D. Diviani).

0167-4889/$ – see front matter © 2012 Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.bbamcr.2012.07.014

ATP-dependent Ca^{2+}-pump SERCA2, promotes SERCA2 activation and reuptake of Ca^{2+} into the sarcoplasmic reticulum, which favors myocyte relaxation [16]. On the other hand, phosphorylation of sarcomeric proteins such as cardiac troponin I (cTnI) and myosin binding protein C (cMyBP-C) influences the dynamics of cardiomyocyte contraction [17–19]. Finally, PKA-mediated regulation of the delayed rectifier potassium channel α subunit (KCNQ1) increases the *IK* current, accelerates repolarization, and shortens ventricular action potential duration [20].

It is now well established that PKA anchoring through AKAPs plays an important role in modulating all these cardiac functions. Pioneer studies using competitor peptides or small molecule compounds that competitively inhibit the interaction between AKAPs and the regulatory subunits of PKA indicated that disruption of PKA anchoring reduced the phosphorylation of cardiac PKA substrates and modified the kinetic of cardiomyocyte contraction in response to β-adrenergic stimulation [21–24]. In recent years, RNA interference and knockout studies have identified several AKAP-based transduction complexes involved in the regulation of calcium cycling and cardiac rhythm. These findings have been reviewed extensively in recent years and will not be discussed further [10,24,25].

In this review article, we will provide an overview of recent literature describing the role of AKAPs in cardiac pathophysiology. Particular emphasis will be given to the adaptive process associated with cardiac hypoxia as well as the remodeling events linked to cardiac hypertrophy and heart failure.

3. Adaptation to hypoxia

Myocardial oxygen concentration needs to be maintained within a narrow range to support cardiac function. During ischemic insult, several adaptive mechanisms are activated within the heart that are aimed at preserving cardiomyocyte function and integrity in conditions of low oxygen supply. In this section we will discuss recent evidence highlighting the role of the anchoring proteins mAKAPβ (AKAP6) and AKAP121 (AKAP1) in coordinating signaling enzymes that control the adaptive response to cardiac hypoxia.

3.1. mAKAPβ (AKAP6)

The transcription factor hypoxia inducible factor 1α (HIF-1α) is a key regulator of the cellular responses to hypoxia. In normoxia, HIF-1α is hydroxylated on two specific proline residues in the oxygen-dependent degradation domain (ODD) by prolyl hydroxylase domain proteins (PHDs) [26,27]. Hydroxylated HIF-1α is subsequently recognized by the von Hippel–Lindau protein (pVHL), which recruits the Elongin C ubiquitin ligase complex, to ubiquitinate HIF-1α and to promote its proteasome-dependent degradation [28,29]. Under hypoxic conditions, PHDs become inactive because of the lack of sufficient oxygen. In parallel ubiquitination of PHD proteins by the E3 ubiquitin ligase *seven in absentia homolog* 2 (Siah2), marks them for subsequent degradation by the proteasome machinery [30]. This allows HIF-1α to escape degradation and to accumulate in the nucleus, where it can dimerize with HIF-1β to promote the transcription of target genes that control adaptive responses to hypoxia including angiogenesis, vascular remodeling, glucose transport, glycolysis, iron transport, cell proliferation, and cell survival [27,30].

Recent findings suggest that the muscle specific anchoring protein mAKAPβ plays a role in cardiomyocyte oxygen homeostasis by organizing a multiprotein complex that controls HIF-1α stability. mAKAPβ has been shown to assemble a signaling complex containing HIF-1α PHD, pVHL and Siah2 in primary cultures of rat neonatal ventricular cardiomyocytes (NVMs) (Fig. 1A) [31]. Under conditions of normal oxygen supply, mAKAPβ-anchored PHD and VHL favor HIF-1α ubiquitylation and degradation. Under hypoxic conditions, however, activation of the E3 ligase Siah2 within the mAKAP complex induces proteasomal degradation of bound PHD. This prevents HIF-1α hydroxylation, ubiquitylation and degradation, and favors HIF-1α-mediated transcription (Fig. 1A) [31]. Therefore, these findings indicate that, by favoring HIF-1α stabilization in response to hypoxia, mAKAPβ enhances the transcriptional activation of HIF-1α regulated genes in cardiomyocytes [31].

mAKAPβ is tethered to the perinuclear membrane of cardiomyocytes through an interaction with Nesprin-1α suggesting that it functions to optimally position HIF-1α in proximity of its site of action inside the nucleus, which may favor rapid transcriptional regulation in response to diminished oxygen availability.

These findings suggest that mAKAPβ/HIF-1α complex could play an important role in promoting cardiomyocyte protection during ischemia arising from partial or complete interruption of coronary blood flow or myocardial hypoxia that develops as a result of a mismatch between myocardial oxygen demand and supply during stress-induced hypertrophy. In agreement with this assumption, the increased mAKAPβ expression associated with cardiac hypertrophy could enhance HIF-1α-mediated transcriptional responses controlling the induction of glycolysis (which maximizes ATP production under hypoxic conditions), the efficiency of mitochondrial respiration and cell survival during ischemia [27].

3.2. AKAP121 (AKAP1)

Mitochondria constitute the major source of energy in the heart and alterations of mitochondrial function and dynamics caused by hypoxia profoundly impact cardiomyocyte survival. A crucial aspect of mitochondrial function is the balance of fusion and fission (fragmentation), which impacts mitochondria morphology and activity [32,33]. In this context, accumulating evidence point to a causative or contributory role for increased mitochondrial fission in cardiomyocyte apoptosis during ischemia [33,34].

At the molecular level, mitochondrial fission is regulated by dynamin-related protein 1 (Drp1), a cytosolic GTPase that is recruited to the surface of mitochondria by the outer membrane protein Fis1 [35,36]. DrpI-mediated GTP hydrolysis provides the driving force required for mitochondrial membrane fission [35,36]. Recent evidence now indicates that the mitochondrial anchoring protein AKAP121 [37] (also known as D-AKAP1 and AKAP1) plays a crucial role in regulating DrpI-mediated fission during hypoxia [38]. AKAP121 controls the balance between mitochondrial fusion and fission by forming a complex with DrpI, by favoring PKA-mediated phosphorylation and inactivation of bound DrpI as well as by limiting the formation of DrpI–Fis1 complexes at the mitochondrial surface through a PKA-independent mechanism that remains to be characterized (Fig. 1B) [38]. Under hypoxic conditions, Siah2 induces AKAP121 ubiquitination and degradation [38]. As a result, the reduced expression of AKAP121 relieves PKA-mediated DrpI inhibition and promotes the formation of DrpI–FisI complexes, resulting in mitochondrial fission and increased apoptosis (Fig. 1B). In line with these findings, recent results indicate that displacement of AKAP121 from mitochondria using competitive peptides promotes mitochondrial dysfunction and induces cardiomyocyte apoptosis in rat hearts [39].

Additional evidence indicates that AKAP121 can influence cell death by favoring PKA-mediated phosphorylation of the pro-apoptotic protein BAD in PC12 and hematopoietic FL5.12 cells [40,41]. This inhibits the interaction between BAD and the anti-apoptotic protein Bcl2, which ultimately causes cytochrome C release from mitochondria, activation of effector caspases and apoptosis [40,41].

Altogether, while the findings described above indicate that AKAP121 plays a crucial role in antagonizing apoptotic cardiomyocyte death in cardiomyocyte cultures, future studies will need to address whether hypoxia-mediated AKAP121 degradation contributes to cardiomyocyte death *in vivo*.

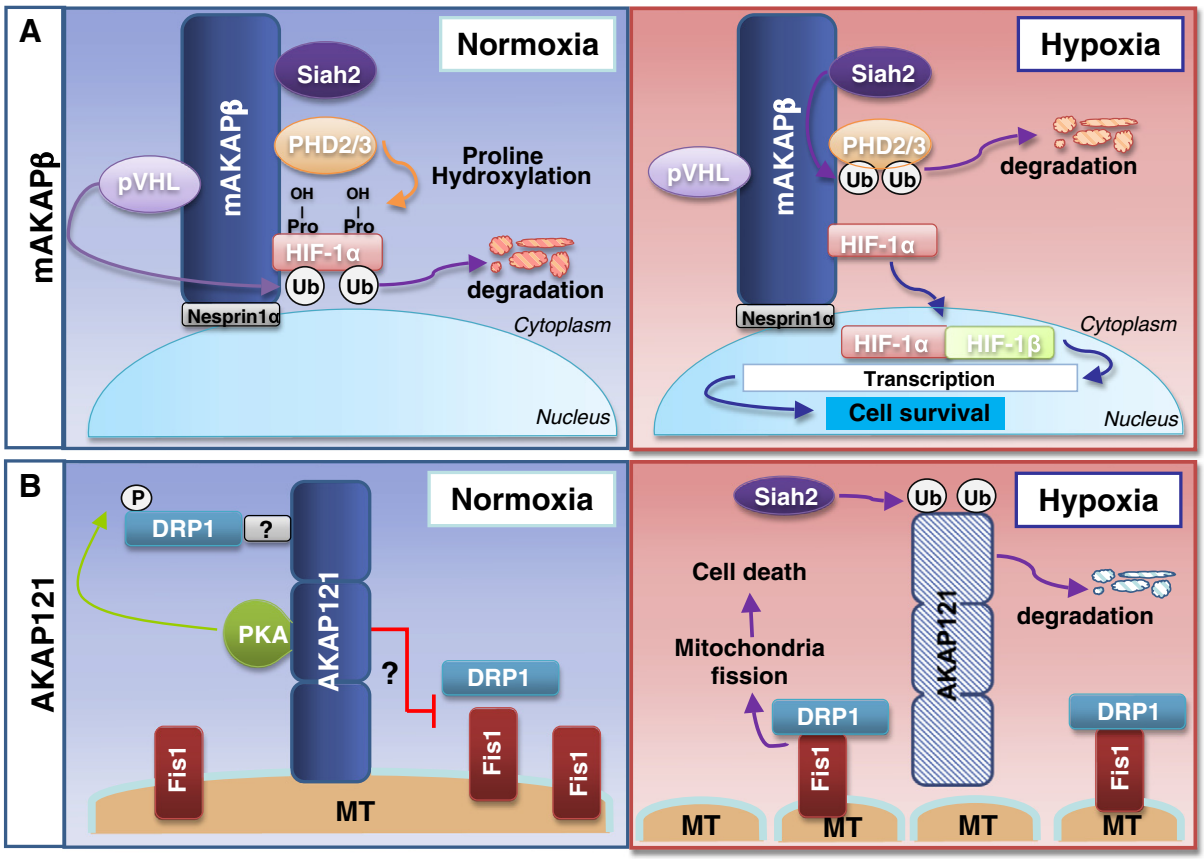

Fig. 1. AKAPs regulating the cardiac adaptive response to hypoxia. A) Regulation of HIF1α-mediated transcription during normoxia and hypoxia by mAKAP. Under normoxic conditions (left panel), HIF-1α is hydroxylated on proline residues (Pro^{402} and Pro^{564}) by PHD and subsequently recognized and ubiquitinated by pVHL. Ubiquitinated HIF-1α is targeted to the proteasome for degradation. Under hypoxic conditions (right panel), PHD is ubiquitinated by Siah2 and targeted to the proteasome. This favors HIF-1α stability, allowing the protein to translocate to the nucleus and forming a heterodimeric complex with HIF-1β. This leads to transcription of proangiogenic, metabolic and antiapoptotic genes that favor cell survival during hypoxia. B) Involvement of AKAP121 in regulation of mitochondria dynamics (fission) during normoxia and hypoxia. Under normoxia (left panel), AKAP121 favors PKA-mediated phosphorylation and inactivation of bound Drpl and inhibits the formation of Drpl–Fis1-containing fission complexes at the mitochondrial surface through a PKA-independent mechanism. Under hypoxia (right panel), Siah2 ubiquitinates AKAP121 and reduces its availability. This favors DRP1–Fis1 interaction, which results in mitochondria fission and cell death.

4. Cardiac hypertrophy

Ventricular cardiac hypertrophy is the primary adaptive response whereby the heart responds to hemodynamic or neurohumoral stresses [2]. It is associated with an increase in cardiomyocyte size, sarcomere reorganization and the activation of an embryonic gene program. Aberrant expression during adult life of fetal proteins involved in cardiac contractility, calcium handling and myocardial energetics leads to maladaptive changes that, on the long-term, alter contractility and induce cardiomyocyte death [6].

Mechanical stress has been proposed to promote cardiomyocyte hypertrophy by activating stretch-sensitive ion channels, integrins, and other structural proteins in a complex network that links the extracellular matrix, the cytoskeleton, the sarcomere and the nucleus [42], as well as by directly activating the angiotensin II receptor in a ligand-independent manner [43]. On the other hand, many neurohumoral stimuli induce cardiac growth by activating G protein coupled receptors (GPCRs) coupled to the hererotrimeric G protein Gq, such as the α1-adrenergic- (α1-ARs), angiotensin-II- (AT1-Rs) and endothelin-1-receptors (ET1-Rs) [44]; or Gs, such as the β-adrenergic receptors (β-ARs) [45].

Evidence collected over the last seven years indicates that AKAPs play a central role in coordinating multiple pro-hypertrophic signaling pathways activated by stress stimuli. In particular, mAKAPβ and AKAP-Lbc have been identified as organizers of multienzyme transduction complexes promoting cardiomyocyte hypertrophy [46–48], whereas AKAP121 has been identified as a negative modulator of stress-induced hypertrophic signaling [49]. These findings are described below.

4.1. mAKAPβ (AKAP6)

The mAKAPβ is a perinuclear anchoring protein that coordinates multiple pathways controlling cardiomyocyte hypertrophy in response to extracellular stimuli including catecholamines, endothelin and cytokines. In this context, RNA interference studies performed in NVMs indicate that mAKAP knockdown significantly reduces hypertrophy and fetal gene expression induced by the activation of endogenous α1- and β-adrenergic receptors (ARs), Endothelin1-receptors (ET1-Rs) and gp130/leukemia inhibitor factor (LIF) receptors [45,46]. At the perinuclear membrane, mAKAPβ recruits a repertoire of multiple signaling enzymes that control hypertrophic gene transcription through at least three mechanisms.

The first, requires the formation of a complex between mAKAPβ, PKA, RyR2, calcineurin Aβ (CaNAβ) and the pro-hypertrophic transcription factor NFATc3 [47]. In response to β-AR activation, anchored PKA promotes RyR2 phosphorylation. The consequent release of Ca^{2+} is proposed to activate bound CaNAβ, which, in turn, dephosphorylates NFATc3 and promotes its nuclear translocation, thus favoring hypertrophic gene transcription (Fig. 2A) [47,50].

The second, requires the interaction with phosphodiesterase 4D3, which, in turn recruits the cAMP-activated guanine nucleotide

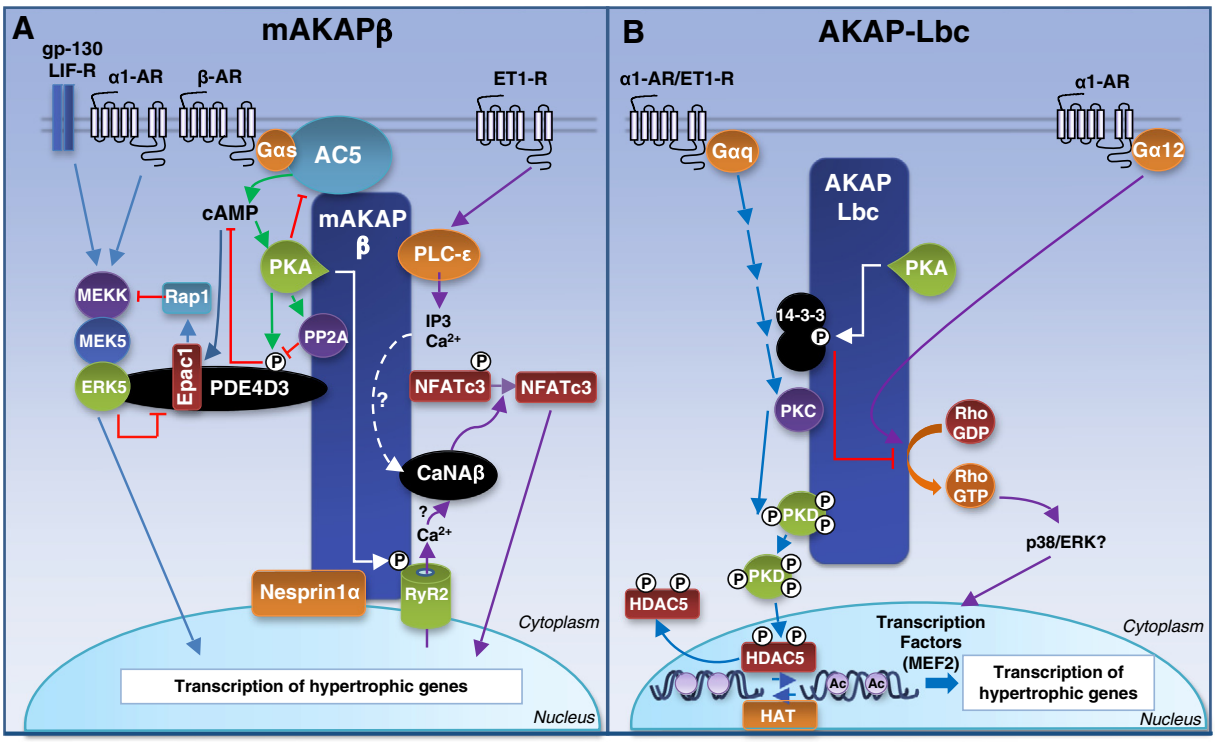

Fig. 2. AKAPs regulating cardiac hypertrophy. A) mAKAPβ assembles a multivalent transduction complex localized at the outer nuclear membrane of cardiomyocytes that integrates hypertrophic signals from multiple membrane receptors including β-ARs, α1-ARs, ET_1-Rs and LIF/gp130-Rs. β-Adrenergic stimulation generates cAMP, through the activation of mAKAPβ-associated AC5. The raise in cAMP concentration activates anchored PKA, leading to the phosphorylation of RyR2 and to an increased release of Ca^{2+} from intracellular stores. This might locally activate mAKAP-bound CaNAβ leading to the dephosphorylation and nuclear translocation of the transcription factor NFATc3 what will induce hypertrophic gene transcription. Anchored PKA can also phosphorylate additional enzymes within the mAKAPβ complex including PDE4D3 and AC5 and PP2A. Phosphorylation of PDE4D3 and AC5 enhances local cAMP degradation and inhibits cAMP synthesis, respectively, whereas phosphorylated PP2A promotes PDE4D3 dephosphorylation favoring cAMP signaling. In response to ET_1-Rs stimulation, mAKAPβ-bound PLC-ε promotes cardiomyocyte hypertrophy *via* an unknown pathway that may involve inositol-1,4,5-triphosphate production, mobilization of local Ca^{2+} stores, and activation of mAKAP-bound CaNAβ. Stimulation of LIF/Gp130-Rs and α1-ARs activates hypertrophic MEK5-ERK5 signaling. mAKAP-anchored ERK5 is proposed to promote cardiomyocyte hypertrophy through the activation of MEF2 and/or through the inhibition of PDE4D3 and the consequent stimulation of the cAMP-PKA-Ryr2-calcineurin-NFATc3 pathway described above. High cAMP concentrations inhibit anchored ERK5 through a feedback mechanism that requires activation of Epac and its effector Rap1. B) The Rho-GEF activity of AKAP-Lbc is enhanced by α_1-ARs through a $G\alpha_{12}$-dependent pathway. Activated RhoA promotes cardiomyocytes hypertrophy through signaling cascades that are yet to be characterized. Possible effector pathways might involve p38 and ERK. Phosphorylation of AKAP-Lbc on serine-1565 by anchored PKA induces the recruitment of 14-3-3, which inhibits the Rho-GEF activity of AKAP-Lbc. AKAP-Lbc also serves a scaffold for PKCη and its downstream effector PKD. Anchored PKCη is activated in response to α_1-ARs and ET_1Rs stimulation and phosphorylates PKD. In turn, PKD translocates to the nucleus where it promotes HDAC5 phosphorylation and nuclear export favoring MEF2-dependent transcription of fetal genes.

exchange factor Epac1, the pro-hypertrophic mitogen-activated protein kinase (MAPK) ERK5 and its upstream activator MEK5 [46]. Activation of gp130 by LIF stimulates MEK5–ERK5 signaling (Fig. 2A). It is not entirely clear how anchored ERK5 favors cardiac hypertrophy. One possibility is that it could favor the phosphorylation and nuclear activation of MEF2 [51]. Alternatively, ERK5 mediated inhibition of PDE4D3 could increase the local concentration of cAMP, and, as a consequence enhance the PKA–Ryr2–calcineurin–NFATc3 pathway described above (Fig. 2A) [47]. High cAMP concentrations inhibit anchored ERK5 through a feedback mechanism that requires activation of Epac and its effector Rap1 (Fig. 2A) [46].

Interestingly, PDE4D3 plays an important role in regulating cAMP levels in the vicinity of the anchoring protein. When cAMP concentrations rise, anchored PKA phosphorylates and activates PDE4D3, which depletes the local concentrations of cAMP and enables reformation of the inactive PKA holoenzyme [46,52]. This implies that the mAKAP complex generates cyclic pulses of PKA activity (Fig. 2A).

The third mechanism whereby mAKAPβ regulates cardiomyocyte hypertrophy involves the recruitment of phospholipase C-ε (PLC-ε). This was mainly demonstrated by RNA silencing experiments showing that disruption of mAKAP–PLC-ε complexes in rat NVMs attenuates endothelin 1-induced hypertrophy [53]. It is currently unknown how anchored PLC-ε promotes hypertrophy but possible mechanisms might involve the local formation of inositol 1,4,5-phosphate, which could mobilize intracellular Ca^{2+} and promote the activation of mAKAPβ-bound CaNAβ and NFATc3 (Fig. 2A). This might lead to NFATc3 dephosphorylation and nuclear translocation, and favor hypertrophic gene transcription.

Collectively, these findings indicate that the mAKAPβ signaling complex coordinates signals emanating from diverse membrane receptors to favor NFAT-dependent hypertrophic gene transcription.

4.2. AKAP-Lbc (AKAP13)

AKAP-Lbc is a cardiac anchoring protein that integrates signals from multiple membrane receptors and conveys them to the activation of transcriptional responses that ultimately regulate the expression of genes associated with hypertrophy [54].

AKAP-Lbc acts as a RhoA selective guanine nucleotide exchange factor (GEF) [55] and serves as a scaffold for multiple signaling enzymes. RhoA activation is mediated by a GEF module constituted of consecutive Dbl (DH) and pleckstrin homology (PH) domains [55]. In cardiomyocytes, AKAP-Lbc Rho-GEF activity is enhanced by α1-ARs through a transduction cascade involving the α subunit of the heterotrimeric G protein G12 (Fig. 2B) [48,55].

Silencing of AKAP-Lbc expression or inhibiting Gα12 function in rat NVMs strongly reduces both α1-AR-mediated RhoA activation and hypertrophic responses [48,56]. This suggests that this anchoring

protein coordinates a pro-hypertrophic transduction pathway activated by the α1-AR that includes Gα12, AKAP-Lbc and RhoA (Fig. 2B). While it is currently unknown which Rho effector molecules mediate the hypertrophic effect of AKAP-Lbc in cardiomyocytes, recent studies performed in cell lines indicate that the anchoring protein can recruit signaling enzymes involved in the activation of different mitogen activated protein kinase (MAPK) pathways [57,58]. In particular, AKAP-Lbc has been shown to assemble a transduction complex containing the scaffold kinase suppressor of Ras (KSR-1), Raf and MEK1/2, that coordinates the activation of ERK1/2 [57]; and to mobilize a signaling unit that includes the RhoA effector PKNα, MLTK, MKK3 and p38α, which promotes RhoA dependent activation of p38α [58]. Given the importance of ERK1/2- and p38α-mediated pathways in regulating cardiomyocyte growth *in vivo* [59,60], it will be crucial to define whether formation of these complexes within the heart can influence the hypertrophic response to stress stimuli.

In addition to its RhoA-mediated effects on cardiomyocyte growth, additional findings demonstrate that AKAP-Lbc also coordinates a protein kinase D (PKD) pathway regulating the expression of fetal genes (Fig. 2B) [56,61]. In particular, AKAP-Lbc can recruit protein kinase Cη (PKCη) and its downstream effector PKD. In response to phenylephrine or endothelin I stimulation, anchored PKCη phosphorylates and activates PKD, which, after being released from the AKAP-Lbc complex, can induce phosphorylation of histone deacetylase 5 (HDAC5) to promote its nuclear export [56]. This is proposed to lead to chromatin derepression and to favor myocyte enhanced binding factor 2 (MEF-2)-dependent transcription of hypertrophic genes (Fig. 2B). Accordingly, downregulation of AKAP-Lbc expression in rat NVMs suppressed the nuclear export of HDAC5 and repressed transcription of the atrial natriuretic factor (ANF) gene, a marker for pathological cardiac hypertrophy [56].

Recent evidence indicates that AKAP-Lbc anchors PKA to favor β-adrenergic receptor-mediated phosphorylation of the heat shock protein 20 (HSP20) in cardiomyocytes [62]. Knowing that PKA-mediated phosphorylation of HSP20 can protect against hypertrophy [63], one could raise the hypothesis that AKAP-Lbc might also coordinate anti-hypertrophic pathways.

Importantly, AKAP-Lbc expression is strongly increased in the heart of mice submitted to neurohumoral or hemodynamic stresses including chronic infusion of phenylephrine [48], angiotensin II and thoracic aortic constriction (A. Appert-Collin and D. Diviani, unpublished observations). In line with these findings, analysis of AKAP-Lbc expression in heart samples from patients with hypertrophic cardiomyopathy revealed a twofold increase in the AKAP-Lbc mRNA content over normal patient control samples [56], suggesting a possible role for the anchoring protein in regulating pathological cardiac remodeling. However, direct *in vivo* evidence for an implication of AKAP-Lbc in cardiac hypertrophy is currently lacking mainly because AKAP-Lbc knockout mice die *in utero* due to heart developmental defects [64]. Future experiments using knock-in and/or tissue specific inducible knockout models will likely solve this problem and elucidate the relative contribution of the multiple signaling pathways activated by the anchoring protein in this heart pathology. One could also speculate that interfering with the ability of AKAP-Lbc to activate RhoA and/or to interact with PKD might represent a potential interesting strategy to reduce cardiac hypertrophy *in vivo*.

4.3. AKAP121 (AKAP1)

Recent evidence suggests that AKAP121 functions as negative regulator of cardiomyocyte hypertrophy. Knockdown of AKAP121 expression by RNA interference in rat NVMs promotes cardiomyocyte growth, whereas its overexpression reduces both basal cardiomyocyte size and the pro-hypertrophic effects of the β-adrenergic agonist isoproterenol [49]. At the molecular level, these effects can be explained by the fact that AKAP121 can sequester and inhibit the phosphatase calcineurin. This affects the downstream activation of the transcription factor NFAT, which requires calcineurin-mediated dephosphorylation in order to translocate from the cytoplasm to nucleus and activate target hypertrophic genes [49]. Consistent with a role of AKAP121 in cardiomyocyte growth inhibition, induction of hypertrophy in rats by aortic banding strongly reduces the ventricular expression of the anchoring protein [39].

Based on these findings, one could postulate that AKAP121 degradation induced by hypoxia (Fig. 1A) would release calcineurin, and, as a consequence promote activation and nuclear translocation of NFAT. This could potentially provide a mechanistic explanation for the well-established stimulatory effect of hypoxia on cardiac hypertrophy. Future studies will need to address whether alterations of the cardiac expression of this anchoring protein can affect the development of cardiac hypertrophy and dysfunction.

5. Heart failure

Heart failure represents the end phenotype of a number of diverse primary pathologies that include hypertension, coronary artery disease, diabetes and valvular diseases. It is associated with diminished cardiomyocyte contractility, cardiomyocyte apoptosis, fibrosis and reduced ejection fraction. Depressed cardiac function causes reflex sympathetic activation [2]. The consequent release of high levels of catecholamines from sympathetic nerve endings promotes desensitization, internalization and degradation of cardiac β-adrenergic receptors, and inhibits downstream PKA signaling [65]. This attenuates the stimulatory effects of PKA on calcium cycling and reduces contractility [65].

The process of β-AR desensitization and internalization is initiated by the G protein-coupled receptor kinase 2 (GRK2), which phosphorylates ligand-occupied receptors thus favoring the binding of β-arrestin and the adaptor protein complex 2 (AP2), two proteins that mediate the recruitment of clathrin [66–68]. Importantly, GRK2 and AP2 can also bind the phosphoinositide 3-kinase γ (PI3Kγ) and promote its recruitment to the plasma membrane in proximity of the β-adrenergic receptor [69]. At this location PI3Kγ promotes local PIP3 production, which favors receptor internalization. It is now shown that PI3Kγ forms a complex with PKA that regulates β-AR signaling and internalization in cardiomyocytes [70]. Under physiological conditions, the activation of PI3Kγ-anchored PKA by the β-AR/cAMP pathway leads to the phosphorylation of PI3Kγ. This inactivates the kinase and tonically inhibits receptor internalization (Fig. 3, left panel) [70]. During heart failure, PI3Kγ is dramatically upregulated, thus limiting the formation of complexes containing PKA. As a result, PI3Kγ escapes PKA-mediated inactivation, decreases cell surface expression of β-ARs and depresses heart function (Fig. 3, right panel). In line with these findings, pharmacological inhibition of PI3Kγ restores β-AR density and improves contractility in failing hearts [70]. This suggests that targeting the PI3Kγ signaling complexes might emerge as a potential therapeutic approach aimed at normalizing myocardial function during heart failure.

While it is clear that downregulation of β-ARs contributes importantly to depressed cardiac function during heart failure, it is of particular importance to determine whether altered AKAP signaling could also contribute to the profound decrease in PKA activation observed at end-stage heart failure. This hypothesis is consolidated by the findings by Zakhary et al., who showed that the interaction between AKAPs and R subunits of PKA is decreased in failing hearts [71]. This might potentially have important functional consequences since experimental disruption of AKAP–PKA complexes in rat NVMs using competitor peptides corresponding to the regulatory subunit-binding domain of AKAPs can decrease PKA-dependent phosphorylation of RyR2, phospholamban, cTnI and cMyBPC, inhibit cAMP-mediated regulation of L-type Ca^{2+} currents, and alter the kinetics of cardiomyocyte contraction in response to β-adrenergic stimulation [21–24]. Based on this experimental evidence, it will be crucial to determine whether individual AKAP complexes are altered during the

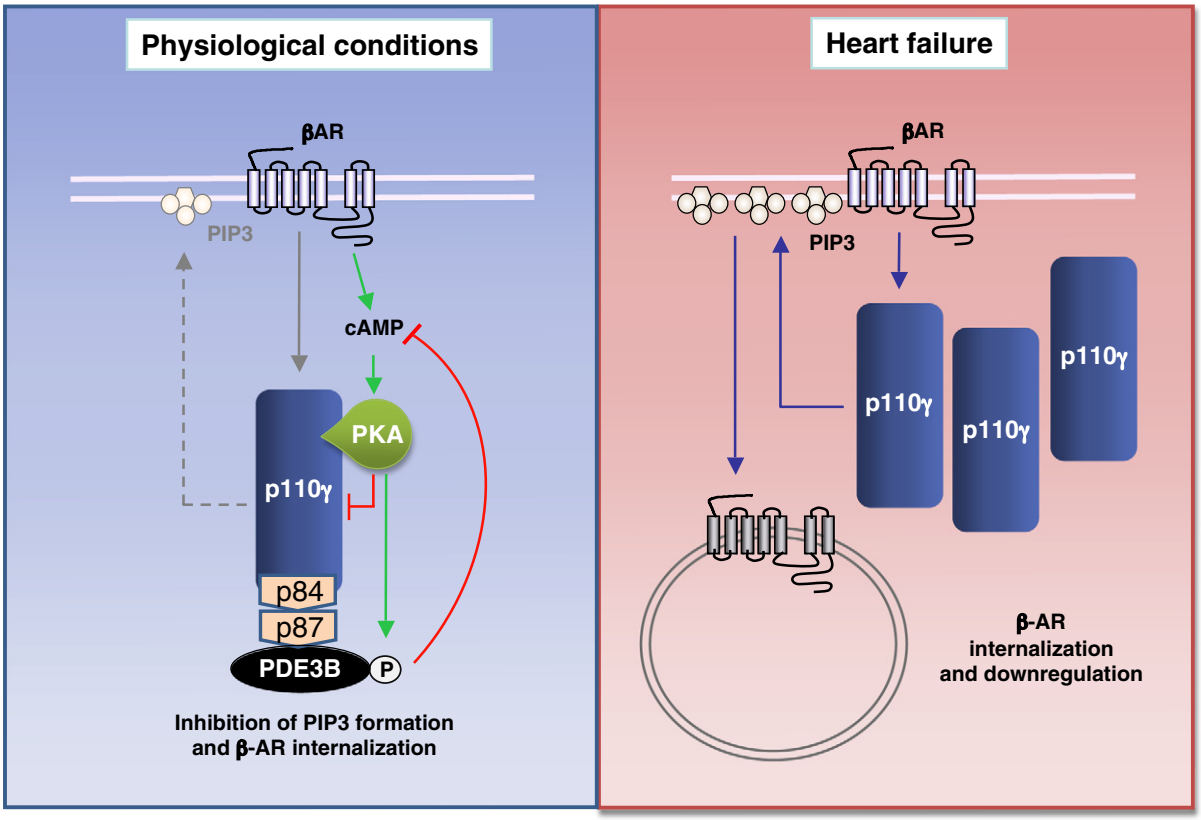

Fig. 3. The role of PI3Kγ–PKA complexes in heart failure. Under physiological conditions, the myocardial PI3Kγ catalytic subunit p110γ forms a complex with the adaptor subunits p84/p87, PDE3B and PKA. Anchored PKA activates PDE3B to enhance cAMP degradation and phosphorylates p110γ to inhibit its lipid kinase activity and maintain β-ARs at the surface of cardiomyocytes. In heart failure, however, upregulation of p110γ uncouples the lipid kinase from PKA. This leads to decreased PKA-mediated p110γ inhibition, which enhances β-ARs internalization and myocardial dysfunction.

transition from cardiac hypertrophy to heart failure and whether this influences the progression of the disease.

6. Conclusions and perspectives

It has become increasingly clear that the aspects of intracellular signaling influencing heart pathophysiology are regulated by cardiac AKAPs, which assemble macromolecular complexes at defined subcellular compartments to provide tight spatiotemporal control to stress signal transmission.

While over the past ten years AKAPs have emerged as key molecular regulators of the cardiac response to injury and stress, more work is required to precisely define the global implication of this family of anchoring proteins in the remodeling process sustained by the stressed heart. In this respect, it will be crucial to determine whether AKAPs could play a role in cardiac fibrosis and in the remodeling of cardiac metabolism, as several experimental evidence now suggests a key role for PKA in the regulation of cardiac fibroblast functions, such as migration and collagen secretion [72], and metabolic processes including glycolysis and oxidative phosphorylation [73–75].

While in past years AKAP function has been mainly studied in isolated cardiomyocytes, a growing number of recent studies now suggest that AKAP-based signaling complexes organize physiologically relevant signaling events also *in vivo*. The generation of inducible cardiac specific knock-out and/or knock-in animal models will certainly expand our current knowledge of how anchoring proteins organize and integrate signaling pathways that influence heart function under both physiological and pathological conditions. They will also provide precious tools to dissect the relevance of individual protein–protein interactions in heart pathophysiology. This information together with the accurate structural analysis of relevant interactive surfaces within AKAP transduction complexes will be instrumental for designing and developing peptides and small molecule inhibitors that could antagonize AKAP-mediated pathological processes in the remodeling heart.

Acknowledgements

This work was supported by grants 3100A0-138289 (to D.D.) of the Fonds National Suisse de la Recherche Scientifique.

References

[1] S.P. Barry, P.A. Townsend, What causes a broken heart—molecular insights into heart failure, Int. Rev. Cell Mol. Biol. 284 (2010) 113–179.
[2] J.A. Hill, E.N. Olson, Cardiac plasticity, N. Engl. J. Med. 358 (2008) 1370–1380.
[3] J.A. Towbin, N.E. Bowles, The failing heart, Nature 415 (2002) 227–233.
[4] K.T. Weber, Y. Sun, J. Diez, Fibrosis: a living tissue and the infarcted heart, J. Am. Coll. Cardiol. 52 (2008) 2029–2031.
[5] T.P. Cappola, Molecular remodeling in human heart failure, J. Am. Coll. Cardiol. 51 (2008) 137–178.
[6] N. Frey, H.A. Katus, E.N. Olson, J.A. Hill, Hypertrophy of the heart: a new therapeutic target? Circulation 109 (2004) 1580–1589.
[7] D.L. Beene, J.D. Scott, A-kinase anchoring proteins take shape, Curr. Opin. Cell Biol. 19 (2007) 192–198.
[8] C.T. Pawson, J.D. Scott, Signal integration through blending, bolstering and bifurcating of intracellular information, Nat. Struct. Mol. Biol. 17 (2010) 653–658.
[9] E.J. Welch, B.W. Jones, J.D. Scott, Networking with AKAPs: context-dependent regulation of anchored enzymes, Mol. Interv. 10 (2010) 86–97.
[10] D. Diviani, Curr. Opin. Pharmacol. 8 (2008) 166–173.
[11] M.G. Gold, B. Lygren, P. Dokurno, N. Hoshi, G. McConnachie, K. Tasken, C.R. Carlson, J.D. Scott, D. Barford, Molecular basis of AKAP specificity for PKA regulatory subunits, Mol. Cell 24 (2006) 383–395.
[12] F.S. Kinderman, C. Kim, S. von Daake, Y. Ma, B.Q. Pham, G. Spraggon, N.H. Xuong, P.A. Jennings, S.S. Taylor, A dynamic mechanism for AKAP binding to RII isoforms of cAMP-dependent protein kinase, Mol. Cell 24 (2006) 397–408.
[13] P. Skroblin, S. Grossmann, G. Schafer, W. Rosenthal, E. Klussmann, Mechanisms of protein kinase A anchoring, Int. Rev. Cell Mol. Biol. 283 (2010) 235–330.

[14] J.T. Hulme, R.E. Westenbroek, T. Scheuer, W.A. Catterall, Phosphorylation of serine 1928 in the distal C-terminal domain of cardiac CaV1.2 channels during beta1-adrenergic regulation, Proc. Natl. Acad. Sci. U. S. A. 103 (2006) 16574–16579.
[15] S.O. Marx, S. Reiken, Y. Hisamatsu, T. Jayaraman, D. Burkhoff, N. Rosemblit, A.R. Marks, PKA phosphorylation dissociates FKBP12.6 from the calcium release channel (ryanodine receptor): defective regulation in failing hearts, Cell 101 (2000) 365–376.
[16] D. Hagemann, R.P. Xiao, Dual site phospholamban phosphorylation and its physiological relevance in the heart, Trends Cardiovasc. Med. 12 (2002) 51–56.
[17] P.P. Chen, J.R. Patel, I.N. Rybakova, J.W. Walker, R.L. Moss, Protein kinase A-induced myofilament desensitization to Ca^{2+} as a result of phosphorylation of cardiac myosin-binding protein C, J. Gen. Physiol. 136 (2010) 615–627.
[18] J.C. Kentish, D.T. McCloskey, J. Layland, S. Palmer, J.M. Leiden, A.F. Martin, R.J. Solaro, Phosphorylation of troponin I by protein kinase A accelerates relaxation and crossbridge cycle kinetics in mouse ventricular muscle, Circ. Res. 88 (2001) 1059–1065.
[19] J.E. Stelzer, J.R. Patel, J.W. Walker, R.L. Moss, Differential roles of cardiac myosin-binding protein C and cardiac troponin I in the myofibrillar force responses to protein kinase A phosphorylation, Circ. Res. 101 (2007) 503–511.
[20] S.O. Marx, J. Kurokawa, S. Reiken, H. Motoike, J. D'Armiento, A.R. Marks, R.S. Kass, Requirement of a macromolecular signaling complex for beta adrenergic receptor modulation of the KCNQ1-KCNE1 potassium channel, Science 295 (2002) 496–499.
[21] M.A. Fink, D.R. Zakhary, J.A. Mackey, R.W. Desnoyer, C. Apperson-Hansen, D.S. Damron, M. Bond, AKAP-mediated targeting of protein kinase a regulates contractility in cardiac myocytes, Circ. Res. 88 (2001) 291–297.
[22] B.K. McConnell, Z. Popovic, N. Mal, K. Lee, J. Bautista, F. Forudi, R. Schwartzman, J.P. Jin, M. Penn, M. Bond, Disruption of protein kinase A interaction with A-kinase-anchoring proteins in the heart *in vivo*: effects on cardiac contractility, protein kinase A phosphorylation, and troponin I proteolysis, J. Biol. Chem. 284 (2009) 1583–1592.
[23] F. Christian, M. Szaszak, S. Friedl, S. Drewianka, D. Lorenz, A. Goncalves, J. Furkert, C. Vargas, P. Schmieder, F. Gotz, K. Zuhlke, M. Moutty, H. Gottert, M. Joshi, B. Reif, H. Haase, I. Morano, S. Grossmann, A. Klukovits, J. Verli, R. Gaspar, C. Noack, M. Bergmann, R. Kass, K. Hampel, D. Kashin, H.G. Genieser, F.W. Herberg, D. Willoughby, D.M. Cooper, G.S. Baillie, M.D. Houslay, J.P. von Kries, B. Zimmermann, W. Rosenthal, E. Klussmann, Small molecule AKAP-protein kinase A (PKA) interaction disruptors that activate PKA interfere with compartmentalized cAMP signaling in cardiac myocytes, J. Biol. Chem. 286 (2011) 9079–9096.
[24] H.H. Patel, L.L. Hamuro, B.J. Chun, Y. Kawaraguchi, A. Quick, B. Rebolledo, J. Pennypacker, J. Thurston, N. Rodriguez-Pinto, C. Self, G. Olson, P.A. Insel, W.R. Giles, S.S. Taylor, D.M. Roth, Disruption of protein kinase A localization using a trans-activator of transcription (TAT)-conjugated A-kinase-anchoring peptide reduces cardiac function, J. Biol. Chem. 285 (2010) 27632–27640.
[25] J.D. Scott, L.F. Santana, A-kinase anchoring proteins: getting to the heart of the matter, Circulation 121 (2010) 1264–1271.
[26] G.L. Semenza, Hypoxia-inducible factor 1 (HIF-1) pathway, Sci. STKE (2007) cm8.
[27] G.L. Semenza, Regulation of oxygen homeostasis by hypoxia-inducible factor 1, Physiology 24 (2009) 97–106.
[28] P. Jaakkola, D.R. Mole, Y.M. Tian, M.I. Wilson, J. Gielbert, S.J. Gaskell, A. Kriegsheim, H.F. Hebestreit, M. Mukherji, C.J. Schofield, P.H. Maxwell, C.W. Pugh, P.J. Ratcliffe, Targeting of HIF-alpha to the von Hippel–Lindau ubiquitylation complex by O_2-regulated prolyl hydroxylation, Science 292 (2001) 468–472.
[29] P.H. Maxwell, M.S. Wiesener, G.W. Chang, S.C. Clifford, E.C. Vaux, M.E. Cockman, C.C. Wykoff, C.W. Pugh, E.R. Maher, P.J. Ratcliffe, The tumour suppressor protein VHL targets hypoxia-inducible factors for oxygen-dependent proteolysis, Nature 399 (1999) 271–275.
[30] K. Nakayama, I.J. Frew, M. Hagensen, M. Skals, H. Habelhah, A. Bhoumik, T. Kadoya, H. Erdjument-Bromage, P. Tempst, P.B. Frappell, D.D. Bowtell, Z. Ronai, Siah2 regulates stability of prolyl-hydroxylases, controls HIF1alpha abundance, and modulates physiological responses to hypoxia, Cell 117 (2004) 941–952.
[31] W. Wong, A.S. Goehring, M.S. Kapiloff, L.K. Langeberg, J.D. Scott, mAKAP compartmentalizes oxygen-dependent control of HIF-1alpha, Sci. Signal. 1 (2008) ra18.
[32] D.C. Chan, Mitochondria: dynamic organelles in disease, aging, and development, Cell 125 (2006) 1241–1252.
[33] D.F. Suen, K.L. Norris, R.J. Youle, Mitochondrial dynamics and apoptosis, Genes Dev. 22 (2008) 1577–1590.
[34] S.B. Ong, S. Subrayan, S.Y. Lim, D.M. Yellon, S.M. Davidson, D.J. Hausenloy, Inhibiting mitochondrial fission protects the heart against ischemia/reperfusion injury, Circulation 121 (2010) 2012–2022.
[35] E. Smirnova, L. Griparic, D.L. Shurland, A.M. van der Bliek, Dynamin-related protein Drp1 is required for mitochondrial division in mammalian cells, Mol. Biol. Cell 12 (2001) 2245–2256.
[36] A. Roux, K. Uyhazi, A. Frost, P. De Camilli, GTP-dependent twisting of dynamin implicates constriction and tension in membrane fission, Nature 441 (2006) 528–531.
[37] L.J. Huang, K. Durick, J.A. Weiner, J. Chun, S.S. Taylor, Identification of a novel dual specificity protein kinase A anchoring protein, D-AKAP1, J. Biol. Chem. 272 (1997) 8057–8064.
[38] H. Kim, M.C. Scimia, D. Wilkinson, R.D. Trelles, M.R. Wood, D. Bowtell, A. Dillin, M. Mercola, Z.A. Ronai, Fine-tuning of Drp1/Fis1 availability by AKAP121/Siah2 regulates mitochondrial adaptation to hypoxia, Mol. Cell 44 (2011) 532–544.
[39] C. Perrino, A. Feliciello, G.G. Schiattarella, G. Esposito, R. Guerriero, L. Zaccaro, A. Del Gatto, M. Saviano, C. Garbi, R. Carangi, E. Di Lorenzo, G. Donato, C. Indolfi, V.E. Avvedimento, M. Chiariello, AKAP121 downregulation impairs protective cAMP signals, promotes mitochondrial dysfunction, and increases oxidative stress, Cardiovasc. Res. 88 (2010) 101–110.
[40] H. Harada, B. Becknell, M. Wilm, M. Mann, L.J. Huang, S.S. Taylor, J.D. Scott, S.J. Korsmeyer, Phosphorylation and inactivation of BAD by mitochondria-anchored protein kinase A, Mol. Cell 3 (1999) 413–422.
[41] A. Affaitati, L. Cardone, T. de Cristofaro, A. Carlucci, M.D. Ginsberg, S. Varrone, M.E. Gottesman, E.V. Avvedimento, A. Feliciello, Essential role of A-kinase anchor protein 121 for cAMP signaling to mitochondria, J. Biol. Chem. 278 (2003) 4286–4294.
[42] J. Sadoshima, S. Izumo, The cellular and molecular response of cardiac myocytes to mechanical stress, Annu. Rev. Physiol. 59 (1997) 551–571.
[43] Y. Zou, H. Akazawa, Y. Qin, M. Sano, H. Takano, T. Minamino, N. Makita, K. Iwanaga, W. Zhu, S. Kudoh, H. Toko, K. Tamura, M. Kihara, T. Nagai, A. Fukamizu, S. Umemura, T. Iiri, T. Fujita, I. Komuro, Mechanical stress activates angiotensin II type 1 receptor without the involvement of angiotensin II, Nat. Cell Biol. 6 (2004) 499–506.
[44] J.H. Brown, D.P. Del Re, M.A. Sussman, The Rac and Rho hall of fame: a decade of hypertrophic signaling hits, Circ. Res. 98 (2006) 730–742.
[45] G.C. Pare, A.L. Bauman, M. McHenry, J.J. Michel, K.L. Dodge-Kafka, M.S. Kapiloff, The mAKAP complex participates in the induction of cardiac myocyte hypertrophy by adrenergic receptor signaling, J. Cell Sci. 118 (2005) 5637–5646.
[46] K.L. Dodge-Kafka, J. Soughayer, G.C. Pare, J.J. Carlisle Michel, L.K. Langeberg, M.S. Kapiloff, J.D. Scott, The protein kinase A anchoring protein mAKAP coordinates two integrated cAMP effector pathways, Nature 437 (2005) 574–578.
[47] J. Li, A. Negro, J. Lopez, A.L. Bauman, E. Henson, K. Dodge-Kafka, M.S. Kapiloff, The mAKAPbeta scaffold regulates cardiac myocyte hypertrophy *via* recruitment of activated calcineurin, J. Mol. Cell. Cardiol. 48 (2010) 387–394.
[48] A. Appert-Collin, S. Cotecchia, M. Nenniger-Tosato, T. Pedrazzini, D. Diviani, The A-kinase anchoring protein (AKAP)-Lbc-signaling complex mediates alpha1 adrenergic receptor-induced cardiomyocyte hypertrophy, Proc. Natl. Acad. Sci. U. S. A. 104 (2007) 10140–10145.
[49] B. Abrenica, M. AlShaaban, M.P. Czubryt, The A-kinase anchor protein AKAP121 is a negative regulator of cardiomyocyte hypertrophy, J. Mol. Cell. Cardiol. 46 (2009) 674–681.
[50] M.S. Kapiloff, N. Jackson, N. Airhart, mAKAP and the ryanodine receptor are part of a multi-component signaling complex on the cardiomyocyte nuclear envelope, J. Cell Sci. 114 (2001) 3167–3176.
[51] R.L. Nicol, N. Frey, G. Pearson, M. Cobb, J. Richardson, E.N. Olson, Activated MEK5 induces serial assembly of sarcomeres and eccentric cardiac hypertrophy, EMBO J. 20 (2001) 2757–2767.
[52] K.L. Dodge, S. Khouangsathiene, M.S. Kapiloff, R. Mouton, E.V. Hill, M.D. Houslay, L.K. Langeberg, J.D. Scott, mAKAP assembles a protein kinase A/PDE4 phosphodiesterase cAMP signaling module, EMBO J. 20 (2001) 1921–1930.
[53] L. Zhang, S. Malik, G.G. Kelley, M.S. Kapiloff, A.V. Smrcka, Phospholipase C epsilon scaffolds to muscle-specific A kinase anchoring protein (mAKAPbeta) and integrates multiple hypertrophic stimuli in cardiac myocytes, J. Biol. Chem. 286 (2011) 23012–23021.
[54] J. Heineke, J.D. Molkentin, Regulation of cardiac hypertrophy by intracellular signalling pathways, Nat. Rev. Mol. Cell Biol. 7 (2006) 589–600.
[55] D. Diviani, J. Soderling, J.D. Scott, AKAP-Lbc anchors protein kinase A and nucleates Galpha 12-selective Rho-mediated stress fiber formation, J. Biol. Chem. 276 (2001) 44247–44257.
[56] G.K. Carnegie, J. Soughayer, F.D. Smith, B.S. Pedroja, F. Zhang, D. Diviani, M.R. Bristow, M.T. Kunkel, A.C. Newton, L.K. Langeberg, J.D. Scott, AKAP-Lbc mobilizes a cardiac hypertrophy signaling pathway, Mol. Cell 32 (2008) 169–179.
[57] F.D. Smith, L.K. Langeberg, C. Cellurale, T. Pawson, D.K. Morrison, R.J. Davis, J.D. Scott, AKAP-Lbc enhances cyclic AMP control of the ERK1/2 cascade, Nat. Cell Biol. 12 (2010) 1242–1249.
[58] L. Cariolato, S. Cavin, D. Diviani, A-kinase anchoring protein (AKAP)-Lbc anchors a PKN-based signaling complex involved in alpha1-adrenergic receptor-induced p38 activation, J. Biol. Chem. 286 (2011) 7925–7937.
[59] I. Kehat, J. Davis, M. Tiburcy, F. Accornero, M.K. Saba-El-Leil, M. Maillet, A.J. York, J.N. Lorenz, W.H. Zimmermann, S. Meloche, J.D. Molkentin, Extracellular signal-regulated kinases 1 and 2 regulate the balance between eccentric and concentric cardiac growth, Circ. Res. 108 (2010) 176–183.
[60] J.M. Streicher, S. Ren, H. Herschman, Y. Wang, MAPK-activated protein kinase-2 in cardiac hypertrophy and cyclooxygenase-2 regulation in heart, Circ. Res. 106 (2010) 1434–1443.
[61] G.K. Carnegie, F.D. Smith, G. McConnachie, L.K. Langeberg, J.D. Scott, AKAP-Lbc nucleates a protein kinase D activation scaffold, Mol. Cell 15 (2004) 889–899.
[62] H.V. Edwards, J.D. Scott, G.S. Baillie, The A-kinase anchoring protein AKAP-Lbc facilitates cardioprotective PKA phosphorylation of Hsp20 on Serine 16, Biochem. J. (2012) (Epub ahead of print), http://dx.doi.org/10.1042/BJ20120570.
[63] H.V. Edwards, J.D. Scott, G.S. Baillie, PKA phosphorylation of the small heat-shock protein Hsp20 enhances its cardioprotective effects, Biochem. Soc. Trans. 40 (2012) 210–214.
[64] C.M. Mayers, J. Wadell, K. McLean, M. Venere, M. Malik, T. Shibata, P.H. Driggers, T. Kino, X.C. Guo, H. Koide, M. Gorivodsky, A. Grinberg, M. Mukhopadhyay, M. Abu-Asab, H. Westphal, J.H. Segars, The Rho guanine nucleotide exchange factor AKAP13 (BRX) is essential for cardiac development in mice, J. Biol. Chem. 285 (2010) 12344–12354.
[65] H.T. Tevaearai, W.J. Koch, Trends molecular restoration of beta-adrenergic receptor signaling improves contractile function of failing hearts, Cardiovasc. Med. 14 (2004) 252–256.
[66] H.A. Rockman, W.J. Koch, R.J. Lefkowitz, Seven-transmembrane-spanning receptors and heart function, Nature 415 (2002) 206–212.

[67] S.A. Laporte, W.E. Miller, K.M. Kim, M.G. Caron, beta-Arrestin/AP-2 interaction in G protein-coupled receptor internalization: identification of a beta-arrestin binging site in beta 2-adaptin, J. Biol. Chem. 277 (2002) 9247–9254.
[68] S.A. Laporte, R.H. Oakley, J.A. Holt, L.S. Barak, M.G. Caron, The interaction of beta-arrestin with the AP-2 adaptor is required for the clustering of beta 2-adrenergic receptor into clathrin-coated pits, J. Biol. Chem. 275 (2000) 23120–23126.
[69] S.V. Naga Prasad, S.A. Laporte, D. Chamberlain, M.G. Caron, L. Barak, H.A. Rockman, Phosphoinositide 3-kinase regulates beta2-adrenergic receptor endocytosis by AP-2 recruitment to the receptor/beta-arrestin complex, J. Cell Biol. 158 (2002) 563–575.
[70] A. Perino, A. Ghigo, E. Ferrero, F. Morello, G. Santulli, G.S. Baillie, F. Damilano, A.J. Dunlop, C. Pawson, R. Walser, R. Levi, F. Altruda, L. Silengo, L.K. Langeberg, G. Neubauer, S. Heyman, G. Lembo, M.P. Wymann, R. Wetzker, M.D. Houslay, G. Iaccarino, J.D. Scott, E. Hirsch, Integrating cardiac PIP3 and cAMP signaling through a PKA anchoring function of p110γ, Mol. Cell 42 (2011) 84–95.
[71] D.R. Zakhary, C.S. Moravec, M. Bond, Regulation of PKA binding to AKAPs in the heart: alterations in human heart failure, Circulation 101 (2000) 1459–1464.
[72] J.S. Swaney, D.M. Roth, E.R. Olson, J.E. Naugle, J.G. Meszaros, P.A. Insel, Inhibition of cardiac myofibroblast formation and collagen synthesis by activation and overexpression of adenylyl cyclase, Proc. Natl. Acad. Sci. U. S. A. 102 (2005) 437–442.
[73] K. Kitamura, K. Kangawa, H. Matsuo, K. Uyeda, Phosphorylation of myocardial fructose-6-phosphate,2-kinase: fructose-2,6-bisphosphatase by cAMP-dependent protein kinase and protein kinase C. Activation by phosphorylation and amino acid sequences of the phosphorylation sites, J. Biol. Chem. 263 (1988) 16796–16801.
[74] J. Deprez, D. Vertommen, D.R. Alessi, L. Hue, M.H. Rider, Phosphorylation and activation of heart 6-phosphofructo-2-kinase by protein kinase B and other protein kinases of the insulin signaling cascades, J. Biol. Chem. 272 (1997) 17269–17275.
[75] S. Papa, A.M. Sardanelli, S. Scacco, V. Petruzzella, Z. Technikova-Dobrova, R. Vergari, A. Signorile, The NADH: ubiquinone oxidoreductase (complex I) of the mammalian respiratory chain and the cAMP cascade, J. Bioenerg. Biomembr. 34 (2002) 1–10.