Actin-associated proteins and cardiomyopathy—the ‘unknown’ beyond troponin and tropomyosin

Elisabeth Ehler1,2

Received: 9 May 2018 / Accepted: 18 May 2018 / Published online: 5 June 2018
© The Author(s) 2018

Abstract
It has been known for several decades that mutations in genes that encode for proteins involved in the control of actomyosin interactions such as the troponin complex, tropomyosin and MYBP-C and thus regulate contraction can lead to hereditary hypertrophic cardiomyopathy. In recent years, it has become apparent that actin-binding proteins not directly involved in the regulation of contraction also can exhibit changed expression levels, show altered subcellular localisation or bear mutations that might lead to hereditary cardiomyopathies. The aim of this review is to look beyond the troponin/tropomyosin mechanism and to give an overview of the different types of actin-associated proteins and their potential roles in cardiomyocytes. It will then discuss recent findings relevant to their involvement in heart disease.

Keywords Actin-binding proteins · Formin · Cytoskeleton · Cardiomyopathy · Intercalated disc

Introduction
Two major different types of cardiomyopathy can be defined in human patients, hypertrophic cardiomyopathy (HCM) and dilated cardiomyopathy (DCM; for review, see Seidman and Seidman 2001). While HCM shows obvious signs of myocyte disarray in conventional histology, the phenotype of DCM is more subtle and can usually only be elucidated by immunohistochemistry and electron microscopy (Pluess and Ehler 2015). The major changes in DCM appear to occur at the intercalated disc, the specialised cell-cell contact between cardiomyocytes. These changes lead to an altered molecular composition and include an increased expression of actin-anchoring proteins (Ehler et al. 2001). In addition, signalling molecules such as PKCalpha are recruited to the intercalated disc (Lange et al. 2016). While about 75% of mutations that lead to hereditary HCM are found in the genes encoding for sarcomeric myosin heavy chain (MYH7) and myosin-binding protein-C (MYBP3; McNally et al. 2013), other components of the myofibrils can be mutated such as the troponins and alpha-tropomyosin (Tardiff 2011). Initially, it was believed that HCM was a disease of the sarcomere. However, with the identification of mutations in more genes that encode for proteins that do not stably associate with myofibrils (Geier et al. 2008), this was probably an over-simplification. Similarly, the hypothesis that hereditary DCM is caused solely by mutations in cytoskeletal proteins had to be abandoned, since mutations in genes that encode for sarcomeric proteins result in this disease phenotype, too (McNally et al. 2013). It may be more the position of the mutation in the molecule or the combination with mutations in other genes that results in a HCM versus a DCM phenotype (McNally and Mestroni 2017; Tardiff 2011).

As far as components of the thin (actin) filaments are concerned, mutations were described for tropomyosin, troponin T, troponin I and troponin C as well as for cardiac actin itself (Hoffmann et al. 2001; Kimura et al. 1997; Olson et al. 1998; Watkins et al. 1995). However, more recently, it was also shown that mutations in actin-interacting proteins that are not directly involved in contraction or its regulation, such as FHOD3, alpha-actinin or filamin C, can cause hereditary cardiomyopathies (Arimura et al. 2013; Girolami et al. 2014;
Tucker et al. 2017; Wooten et al. 2013). These reports prompted the writing of this review on actin and its associated proteins beyond the sarcomere.

Actin is a highly conserved eukaryotic protein that exists as six distinct isoforms: alpha-cardiac, alpha-skeletal, alpha-smooth muscle, beta-cytoplasmic, gamma-cytoplasmic and gamma-smooth muscle actin (Vandekerckhove and Weber 1978). Actin monomers (G-actin) can associate to form filaments (F-actin; see Fig. 1) that have the appearance of two helically entwined pearl strings (Hanson and Lowy 1963). However, this is an energetically unfavourable process, which is massively enhanced by factors that promote actin filament formation such as the Arp2/3 complex or members of the formin family (Chesarone and Goode 2009). Once filaments are formed, they can be stabilised laterally via the association of tropomyosin in one of its numerous isoforms (Gunning et al. 2015). Based on their distinct dynamics, the ends of an actin filament are termed plus end (where incorporation of new actin monomers happens; also called barbed end based on the decoration with myosin heads) and minus end (also called pointed end, where actin monomers are lost in the process of treadmilling). These ends can be protected by the association of capping proteins such as CapZ at the barbed end or tropomodulin and leiomodin at the pointed end (Fig. 2). In addition, actin filaments can be crosslinked to meshworks or bundled to parallel filaments and there are severing proteins that lead to their disassembly (for a landmark review on actin-binding proteins, see Pollard and Cooper 1986, and for a more recent review, see dos Remedios et al. 2003).

Adult cardiomyocytes mainly express the alpha-cardiac actin isoform, which is found almost exclusively in the myofibrils, although cytoplasmic actin isoforms are expressed at very low levels and can be detected in the vicinity of membranes and at the Z-disc (Benz et al. 2013; Dwyer et al. 2012; Kee et al. 2009; Tondeleir et al. 2009, Fig. 2). In the early embryonic heart, alpha-cardiac and alpha-smooth muscle actin are co-expressed in the same thin filaments (Ehler et al. 2004). Confocal microscopy suggests that while the length of the cardiac actin filaments is determined quite early, there exists a population of actin filaments that extends beyond the I-band, which may be mainly composed of alpha-smooth muscle actin (Ehler et al. 2004). Currently, it is unknown whether there are mixed actin filament populations and whether the length-determining factor is functional rather than molecular, since tropomodulin and leiomodin are absent at the pointed ends or not expressed at this developmental stage (Ehler et al. 2004; Tsukada et al. 2010). Similar to other foetal marker genes, an upregulation of alpha-smooth and even alpha-skeletal muscle actin can be detected in hypertrophic cardiomyopathy (Copeland et al. 2010; Suurmeijer et al. 2003). An inbred mouse strain, the Balb/c mouse, also has a higher expression level of alpha-skeletal actin and shows increased contractility (Hewett et al. 1994).

**Actin filament assembly and maintenance in cardiomyocytes**

As mentioned above, since actin filament assembly is an inefficient process, there is a need for factors that might promote it, especially as the half-life of actin and its associated proteins in a cardiomyocyte range from 3 to 10 days (Martin 1981).
The cell migration field has pioneered this research and two main basic ways were identified: (1) filament assembly by the Arp2/3 complex of proteins, which tends to support the formation of filaments at an angle to the mother filaments and (2) filament assembly by members of the formin family, which promote the formation of linear filaments (Chesarone and Goode 2009). Not much is known about Arp2/3 in the heart, but in the skeletal muscle, a role for an Arp2/3 family member, Arpc5L, was shown for the coordination between gamma-actin filaments, the desmin cytoskeleton and nuclear positioning (Roman et al. 2017). More and more of the 15 members of the formin family are characterised as having a role in the heart (Li et al. 2011; Rosado et al. 2014; Taniguchi et al. 2009; reviewed in Randall and Ehler 2014).

Knockout mice for the formin Daam1 mainly reveal a more general role in heart morphogenesis with a noncompaction phenotype and septal defects (Ajima et al. 2015; Li et al. 2011). This is especially interesting, since a recent report showed a potential association of a deletion of a DAAM1 copy with congenital heart disease (Bao et al. 2012). Myofibrils are assembled, but are disorganised, and there may be a problem with their maintenance (Ajima et al. 2015). The major phenotype is seen at the intercalated discs, where cardiomyocyte attachment is severely impaired (Ajima et al. 2015). This is in agreement with localisation data for Daam1 close to the plasma membrane and its potential role in the Wnt effector Dishevelled and thus the planar cell polarity signalling pathway (Li et al. 2011). The phenotype is more severe in Daam1-Daam2 double knockout mice, suggesting a certain redundancy between these proteins.

The formin FHOD3 seems to play a role in early heart development and subsequently in myofibril maintenance. FHOD3 knockout mice did not survive beyond E12.5 and showed hypokinetic ventricles with myofibrillar disarray and Z-disc malformations (Kan-o et al. 2012a). However, a conditional knockout of FHOD3 expression in adult mice did not lead to a lethal phenotype, but just to a mild impairment of cardiac function (Ushijima et al. 2018). Experiments with knockdown of FHOD3 expression in cultured cardiomyocytes also demonstrated a failure to maintain myofibrils and reduced expression levels in samples from human heart failure patients (Iskratsch et al. 2010). Currently, the exact role of FHOD3 in cardiomyocytes is as unclear as its subcellular localisation. We and others detected FHOD3 exclusively at the Z-discs of isolated adult cardiomyocytes and in adult heart tissue from mice and humans (Iskratsch et al. 2010; Rosado et al. 2014), which would fit well with a role as barbed-end facilitator of actin assembly, while others have reported a broader localisation, which overlaps the A-band (Kan-o et al. 2012b). In our hands, this kind of FHOD3 localisation is only detected in the embryonic heart and in cultured neonatal rat cardiomyocytes that are adapting to life in two dimensions in a culture dish (Iskratsch et al. 2010). On the other hand, a recently identified interaction between FHOD3 and MyBP-C, which associates with a subset of the myosin heads, favours the A-band localisation (Matsuyama et al. 2018). Patients with mutations...
in the FHOD3 gene can develop HCM or DCM (Arimura et al. 2013; Wooten et al. 2013). Potentially, FHOD3 is not firmly integrated into the sarcomere and exerts its role by influencing MyBP-C, which is a regulator of the thick filament on-off state (Kampourakis et al. 2014), or it may affect the ratio of available actin monomers. This could explain its detrimental effect on the activation of the transcription factor SRF in the case of the DCM mutant (Arimura et al. 2013).

What happens at the anchorage sites of the myofibrils, the intercalated discs?

As mentioned above, the major subcellular changes observed in DCM occur at the intercalated disc. Both in mouse models for this disease and in human DCM samples, we observed increased expression of all proteins involved in anchoring of actin filaments (i.e. the myofibrils in the cardiomyocytes): the transmembrane cadherins, and at the cytoplasmic face the catenins and N-RAP (Ehler et al. 2001; Pluess et al. 2015). The increased width of signal for these proteins that was seen at the intercalated disc by confocal microscopy was due to a higher degree of membrane convolution, as demonstrated by ultrastructural analysis (Wilson et al. 2014). Analysis of the actin signal using the F-actin stain phalloidin in 0.25-μm-thick cryosections also revealed a higher intensity at the intercalated disc in mouse models for DCM, suggesting that the increased amount of actin-anchoring proteins mirrors an increased presence of filamentous actin there (Ehler et al. 2001). Currently, it is not known which protein is involved in generating more filamentous actin at the intercalated disc, but the observation that the formin FHOD1 locates to this subcellular domain (Al Haj et al. 2015) and its signal is also increased in DCM (Dwyer et al. 2014) makes it a promising candidate. FHOD1 was thought to be unable to promote the formation of actin filaments and to act just as an actin capper and actin bundler (Schönichen et al. 2013). However, FHOD1 participates in the nucleation of actin filaments from early integrin clusters in fibroblasts and is associated with integrins in cardiomyocytes (Al Haj et al. 2015; Iskratsch et al. 2013). Recent evidence also shows that FHOD1’s actin polymerising activity depends on the actin isoform and that while it is inactive with sarcomeric actins (which most people use for in vitro polymerisation assays), it does promote filament formation with cytoplasmic actin isoforms (Patel et al. 2018). Filamentous actin leading up from the transitional junction to the intercalated disc does not seem to contain alpha-cardiac actin (Bennett et al. 2006) and may well be composed of cytoplasmic actins (Benz et al. 2013). Thus, FHOD1 could be an important controlling factor. FHOD1 at the intercalated disc is in an active state, since it can be stained with an antibody against a phosphorylated epitope at T1141. However, it remains to be shown whether it indeed plays a role in excessive actin filament synthesis in DCM (Dwyer et al. 2014).

What happens at the ends of the thin filaments?

Actin-capping proteins determine the length of thin filaments in healthy cardiomyocytes both at the barbed end at the Z-disc and at the pointed end near the inner edges of the H-zone (reviewed by Dwyer et al. 2012; Fowler and Dominguez 2017).

CapZ binds to the barbed ends of thin filaments (Casella et al. 1997) and its dynamics in myocytes is increased by exercise and during hypertrophy (Lin et al. 2013; Lin et al. 2016). Among the signalling pathways that affect CapZ dynamics are PIP2 (phosphatidylinositol-4,5 bisphosphate), phosphorylation by PKC (protein kinase C) and acetylation (Hartman et al. 2009; Lin et al. 2016). CapZ transgenic hearts that express reduced amounts of CapZ are protected against ischemia-reperfusion injury and show alterations in PKC signalling (Yang and Pyle 2012). In addition to its adaptive dynamic behaviour upon cardiomyocyte stress, CapZ also interacts with classical stress signals such as the co-chaperone BAG3 and the small heat shock protein Hsc70 (Hishiya et al. 2010).

Gain of function (overexpression of tropomodulin) and loss of function experiments (interfering with tropomodulin binding) have shown that the tight control of thin filament length at its pointed end is crucial for a healthy cardiomyocyte (Fritz-Six et al. 2003; Gregorio et al. 1995; Sussman et al. 1998) and its pointed end is crucial for a healthy cardiomyocyte (Fritz-Six et al. 2003; Gregorio et al. 1995; Sussman et al. 1998) and its dynamics in myocytes is increased by exercise and during hypertrophy (Lin et al. 2013; Lin et al. 2016). Among the signalling pathways that affect CapZ dynamics are PIP2 (phosphatidylinositol-4,5 bisphosphate), phosphorylation by PKC (protein kinase C) and acetylation (Hartman et al. 2009; Lin et al. 2016). CapZ transgenic hearts that express reduced amounts of CapZ are protected against ischemia-reperfusion injury and show alterations in PKC signalling (Yang and Pyle 2012). In addition to its adaptive dynamic behaviour upon cardiomyocyte stress, CapZ also interacts with classical stress signals such as the co-chaperone BAG3 and the small heat shock protein Hsc70 (Hishiya et al. 2010).

Gain of function (overexpression of tropomodulin) and loss of function experiments (interfering with tropomodulin binding) have shown that the tight control of thin filament length at its pointed end is crucial for a healthy cardiomyocyte (Fritz-Six et al. 2003; Gregorio et al. 1995; Sussman et al. 1998) and its dynamics in myocytes is increased by exercise and during hypertrophy (Lin et al. 2013; Lin et al. 2016). Among the signalling pathways that affect CapZ dynamics are PIP2 (phosphatidylinositol-4,5 bisphosphate), phosphorylation by PKC (protein kinase C) and acetylation (Hartman et al. 2009; Lin et al. 2016). CapZ transgenic hearts that express reduced amounts of CapZ are protected against ischemia-reperfusion injury and show alterations in PKC signalling (Yang and Pyle 2012). In addition to its adaptive dynamic behaviour upon cardiomyocyte stress, CapZ also interacts with classical stress signals such as the co-chaperone BAG3 and the small heat shock protein Hsc70 (Hishiya et al. 2010).

What happens at the Z-discs?

Mutations in several Z-disc proteins are associated with a HCM phenotype (Bos and Ackerman 2010). For example, missense mutations in the gene ACTN2 were described, which encodes the actin-crosslinking protein alpha-actinin,
the marker protein for Z-discs (Chiu et al. 2010). Thorough molecular characterisation of these mutations is still under way, but first results indicate that at least in the case of A119T and G111V mutations, the dynamic behaviour of alpha-actinin and its readiness to incorporate into the Z-disc may be affected (Haywood et al. 2016). A second major Z-disc-associated actin-binding protein that was shown to cause cardiomyopathy when mutated is filamin C. Missense mutations of filamin C cause familial restrictive cardiomyopathy and lead to a loss of filamin C signal at the Z-disc (Tucker et al. 2017). Truncating variants of filamin C and its co-chaperone BAG3 are associated with DCM (Janin et al. 2017). Interestingly, BAG3 stimulates filamin transcription and also spatially regulates mTORC1 signalling to simultaneously induce autophagy of damaged filamin and activate protein synthesis upon mechanical stress in cardiomyocytes (Kathage et al. 2017). Again, these results indicate that actin-associated proteins are not just static glue at their respective sites but closely interweave with signalling pathways that are relevant for the cardiomyocyte stress response.

Proteins involved in actin filament turnover and their role in cardiac disease and repair

Cofilin-2 is a member of the ADF/cofilin family of proteins that acts preferentially at the pointed ends of actin filaments and increases the off-rate by 30-fold (for a review, see dos Remedios et al. 2003). In cultured cardiomyocytes, cofilin-2 was shown to localise towards the M-band region of the sarcomeres, where the pointed ends are found (Kremneva et al. 2014). In a healthy cardiomyocyte, cofilin-2 should not affect the structure of thin filaments too much, since the pointed ends are protected by tropomodulin or leiomodin and cofilin’s depolymerising activity is known to be inhibited by the presence of tropomyosins (dos Remedios et al. 2003). However, when cofilin-2 expression is knocked down in cultured cardiomyocytes, a marked elongation of thin filaments is observed and proper I-band striations are lost (Kremneva et al. 2014). In a mouse model for DCM, the calsarcin knockout mouse, cofilin-2 expression was increased due to a decrease in miRNA miR-301a expression and was subsequently shown to be a direct target for this miRNA (Rangrez et al. 2017). Cofilin-2 function is regulated by phosphorylation, and fasudil, an inhibitor of ROCK (Rho kinase), which has a protective effect against cardiac dysfunction, prevents its phosphorylation and promotes the organisation of actin filaments (Lai et al. 2017). In human idiopathic DCM, aggregates of cofilin-2 in its phosphorylated state were detected in the heart samples of patients (Subramanian et al. 2015). To model reduced cofilin-2 activity, a heterozygous cardiac specific cofilin-2 knockout mouse was generated, which expressed only 40% of cofilin-2 compared to wild-type littermates. These mice displayed dilation and wall thinning of the left ventricle (Subramanian et al. 2015). The reduced contractile function was attributed to disorganised sarcomeres in the heterozygous cofilin-2 mice (Subramanian et al. 2015). These data suggest that cofilin-2 has a regulatory role also in cardiomyocytes and that its expression must be tightly controlled to prevent cardiomyopathy.

Profilin is a protein that sequesters actin monomers and governs their ATP-associated state, leading to a higher affinity for the barbed ends (for a review, see dos Remedios et al. 2003). Interestingly, the barbed ends are classically assumed to be the major site of activity of formins. Formins are characterised by two formin homology (FH) domains, FH1 and FH2. The FH2 domains of two formins dimerise into a doughnut-like structure that forms the business end for formin-promoted actin assembly (Goode and Eck 2007), while the FH1 domain interacts with profilin and may help to shunt profilin-bound actin monomers to the neighbouring FH2 domain. Since profilin dissociates from actin in the presence of PIP and PIP2, the environment of the Z-disc, which is enriched in PIP2 (Pyle et al. 2006; Ribeiro et al. 2014), would be an ideal location to release the actin from profilin and make it available for polymerisation. A recent study has demonstrated that the expression of profilin is increased in a variety of rodent models for hypertrophy in situ and in vitro, but decreased in end-stage heart failure patients (Kooij et al. 2016). In vivo experiments in Drosophila showed that overexpression of profilin leads to longer thin filaments than in control strains and results in a functional phenotype resembling dilated cardiomyopathy (Kooij et al. 2016). Knockdown of profilin expression in cultured cardiomyocytes prevented their hypertrophic response, probably by impaired activation of ERK1/ERK2 signalling (Kooij et al. 2016). In conclusion, profilin appears likely to be crucial for hypertrophic growth in the heart, potentially by delivering actin monomers for assembly by members of the formin family.

Another small actin monomer-binding protein, thymosin beta 4 (Tbeta4), has recently entered the limelight by its ability to enhance cardiac repair in the adult heart following injury (Smart et al. 2011). Tbeta4 was administered to the mice by intraperitoneal injection and somehow seemed to activate a population of stem cells in the epicardial surface of the heart that differentiated into blood vessels but also to a much lower extent into cardiomyocytes. The exact role of Tbeta4 in cardiomyocytes is somewhat unclear at the moment, since Tbeta4 knockout mice had no cardiac phenotype (Banerjee et al. 2012). Therefore, its contribution to improved cardiac repair may be mainly due to its enhancement of vascularisation of the injured heart. On the other hand, another group reported shorter sarcomere length, expression of shorter titin isoforms and a limited contractile reserve in their Tbeta4 knockout mouse.
knockout mice, suggesting that Tbeta4 may be somehow involved in the regulation of alternative splicing of titin, potentially via RBM20 (Guo et al. 2012; Smart et al. 2017).

Concluding remarks

With the advance of next-generation sequencing, it can be expected that many more point mutations will be identified in actin-associated proteins in patients with cardiomyopathy, whose contribution to the observed functional phenotypes will have to be validated. However, it is obvious that the actin cytoskeleton is dynamic and that subtle changes that affect this dynamics and its amount will alter the function of a cardiomyocyte.

Acknowledgements

I would like to thank the collaborators past and present for their hard work and fruitful discussions.

Funding

The research in my laboratory was financially supported by the British Heart Foundation and the Medical Research Council.

Compliance with ethical standards

Conflict of interest

Elisabeth Ehler declares that she has no conflict of interest.

Ethical approval

This article does not contain any studies with human participants or animals performed by the author.

Open Access

This article is distributed under the terms of the Creative Commons Attribution 4.0 International License (http://creativecommons.org/licenses/by/4.0/), which permits unrestricted use, distribution, and reproduction in any medium, provided you give appropriate credit to the original author(s) and the source, provide a link to the distribution, and reproduction in any medium, provided you give appropriate credit to the original author(s) and the source, provide a link to the appropriate version of this article or to the original article, and the original article was published in this journal.

References

Ajima R, Bisson JA, Helt JC, Nakaya MA, Habas R, Tessarollo L, He X, Morrissey EE, Yamaguchi TP, Colten ED (2015) DAAM1 and DAAM2 are co-required for myocardial maturation and sarcomere assembly. Dev Biol 408:126–139

Al Haj A, Mazur AJ, Radaszkiewicz K, Radaszkiewicz T, Makowiecka A, Stopchinski BE, Schonichen A, Geyer M, Mammherz HG (2015) Formin distribution in cardiomyocyte: FHOD1 is a component of intercalated disc and costamere. Eur J Cell Biol 94:101–113

Arimura T, Takeya R, Ishikawa T, Yamano T, Matsuo A, Tsutsumi T, Nomura T, Sumimoto H, Kimura A (2013) Dilated cardiomyopathy-associated FHOD3 variant impairs the ability to induce activation of transcription factor serum response factor. Circ J 77:2990–2996

Banerjee J, Zhang J, Moore-Morris T, Lange S, Shen T, Dalton ND, Gu Y, Peterson KL, Evans SM, Chen J (2012) Thymosin beta 4 is dispensable for murine cardiac development and function. Circ Res 110: 456–464

Bao B, Zhang L, Hu H, Yin S, Liang Z (2012) Deletion of a single-copy DAAM1 gene in congenital heart defect: a case report. BMC Med Genet 13:63

Bennett PM, Maggs AM, Baines AJ, Pinder JC (2006) The transitional junction: a new functional subcellular domain at the intercalated disc. Mol Biol Cell 17:2091–2100

Benz PM, Merkel C, Offner K, Abesser M, Ulrich M, Fischer T, Bayer B, Wagner H, Gambaryan S, Ursitti JA, Adham IM, Linke WA, Feller SM, Fleming I, Renne T, Franzis S, Unger A, Schuh K (2013) Mena/VASP and alphah-Spectrin complexes regulate cytoplasmic actin networks in cardiomyocytes and protect from conduction abnormalities and dilated cardiomyopathy. Cell Commun Signal 11:56

Bos JM, Ackerman MJ (2010) Z-disc genes in hypertrophic cardiomyopathy: stretching the cardiomyopathies? J Am Coll Cardiol 55:1136–1138

Casella JF, Craig SW, Maack DJ, Brown AE (1987) Cap Z(36/32), a barbed end actin-capping protein, is a component of the Z-line of skeletal muscle. J Cell Biol 105:371–379

Chereau D, Boczkowska M, Skwarek-Maruszewska A, Fujiwara I, Hayes DB, Rebowski G, Lappalainen P, Pollard TD, Dominguez R (2008) Leiomodin is an actin filament nucleator in muscle cells. Science 320:239–243

Chesarone MA, Goode BL (2009) Actin nucleation and elongation factors: mechanisms and interplay. Curr Opin Cell Biol 21:28–37

Chiu C, Bagnall RD, Ingles J, Yeates L, Kennerson M, Donald JA, Jormakka M, Lind JM, Sensarain C (2010) Mutations in alpha-actinin-2 cause hypertrophic cardiomyopathy: a genome-wide analysis. J Am Coll Cardiol 55:1127–1135

Copeland O, Nowak KJ, Laing NG, Ravenscroft G, Messer AE, Bayliss CR, Marston SB (2010) Investigation of changes in skeletal muscle alpha-actin expression in normal and pathological human and mouse hearts. J Muscle Res Cell Motil 31:207–214

dos Remedios CG, Chhabra D, Kekic M, Dedova IV, Tsubakihara M, Berry DA, Nosworthy NJ (2003) Actin binding proteins: regulation of cytoskeletal microfilaments. Physiol Rev 83:433–473

Dwyer J, Iskratsch T, Ehler E (2012) Actin in striated muscle: recent insights into assembly and maintenance. Biophys Rev 4:17–25

Dwyer J, Pluess M, Iskratsch T, dos Remedios CG, Ehler E (2014) The formin FHOD1 in cardiomyocytes. Anat Rec (Hoboken) 297:1560–1570

Ehler E, Horovits R, Zappinger C, Price RL, Perriard E, Len M, Caroni P, Sussman M, Eppenberger HM, Perriard JC (2001) Alterations at the intercalated disk associated with the absence of muscle LIM protein. J Cell Biol 153:763–772

Ehler E, Fowler VM, Perriard JC (2004) Myofibrillogenesis in the developing chicken heart: role of actin isoforms and of the pointed end capping protein tropomodulin during thin filament assembly. Dev Dyn 229:745–755

Fowler VM, Dominguez R (2017) Tropomodulins and leiomodins: actin pointed end caps and nucleators in muscles. Biophys J 112:1742–1760

Fritz-Six KL, Cox PR, Fischer RS, Xu B, Gregorio CC, Zoghbi HY, Fowler VM (2003) Aberrant myofibril assembly in tropomodulin1 null mice leads to aborted heart development and embryonic lethality. J Cell Biol 163:1033–1044

Geier C, Gehmlich K, Ehler E, Hassfeld S, Perrot A, Hayess K, Cardin N, Wenzel K, Erdmann B, Krackhardt F, Posch MG, Osterziel KJ, Bublak A, Nagele H, Bruckdorfer K, Dietz R, Chien KR, Spuler S, Fürst DO, Nurnberg P, Ozcelik C (2008) Beyond the sarcomere: CSR62 mutations cause hypertrophic cardiomyopathy. Hum Mol Genet 17:2753–2765

Girolami F, Iascone M, Tomberli B, Bardi S, Benelli M, Marseglia G, Sana ME, Basso C, Marziliano N, Merlini PA, Formaro A, Cecchi F, Torricelli F, Olivotti I (2014) Novel alpha-actinin 2 variant associated with familial hypertrophic
cardiomyopathy and juvenile atrial arrhythmias: a massively parallel sequencing study. Circ Cardiovasc Genet 7:741–750
Goode BL, Eck MJ (2007) Mechanism and function of formins in the control of actin assembly. Annu Rev Biochem 76:593–627
Gregorio CC, Weber A, Bondad M, Pennise CR, Fowler VM (1995) Requirement of pointed-end capping by tropomodulin to maintain actin filament length in embryonic chick cardiac myocytes. Nature 377:83–86
Gunning PW, Hardeman EC, Lappalainen P, Mulvihill DP (2015) Tropomyosin—master regulator of actin filament function in the cytoskeleton. J Cell Sci 128:2965–2974
Guo W, Schaler S, Greaser M, Radke MH, Liss M, Govindarajan T, Maatz H, Schulz H, Li S, Parrish AM, Daussaite V, Vakeel P, Klaassen S, Gerull D, Thierfelder L, Regitz-Zugrosek V, Hacker TA, Saue KW, Dec GW, Ellinor PT, MacRae CA, Spallke B, Fischer R, Perrot A, Ozcelik C, Saar K, Hubner N, Gotthardt M (2012) RBM20, a gene for hereditary cardiomyopathy, regulates titin splicing. Nat Med 18:766–773
Hanson J, Lowy J (1963) The structure of F-actin and of actin filaments isolated from muscle. J Mol Biol 6:46–60
Hartman TJ, Martin JL, Solaro RJ, Samarel AM, Russell B (2009) CapZ dynamics are altered by endothelin-1 and phenylephrine via PIP2-and PKC-dependent mechanisms. Am J Physiol Cell Physiol 296: C1034–C1039
Haywood NJ, Wolny M, Rogers B, Trinh CH, Shuping Y, Edwards TA, Hishiya A, Kitazawa T, Takayama S (2010) BAG3 and Hsc70 interact with actin filament length in embryonic chick cardiac myocytes. Nature 467:83–87
Hewett TE, Grupp IL, Grupp G, Robbins J (1994) Alpha-skeletal actin is required for pointed-end capping by tropomodulin to maintain myofibrillar integrity under mechanical stress. Circ Res 74:740–746
Hishiya A, Kitazawa T, Takayama S (2010) BAG3 and Hsc70 interact with actin capping protein CapZ to maintain myofibrillar integrity under mechanical stress. Circ Res 107:1220–1231
Hoffmann B, Schmidt-Trapsh H, Perrot A, Osterziel KJ, Gessner R (2001) First mutation in cardiac troponin C, L29Q, in a patient with hypertrophic cardiomyopathy. Hum Mutat 17:524
Iskrachts T, Lange S, Dwyer J, Kho AL, dos Remedios C, Ehler E (2010) FHOD3 is regulated by CK2 phosphorylation and promotes myofibril maintenance. J Cell Biol 191:1159–1172
Iskrachts T, Yu CH, Mathur A, Dwyer J, Liu S, Stévenin V, Hone J, Ehler E, Sheetz M (2013) FHOD1 is needed for directed forces and adhesion dynamics during cell spreading and migration. Dev Cell 27: 545–559
Janin A, Nguyen K, Habib G, Dauphin C, Chanavat V, Bouvagnet P, Eschalier R, Streichenberger N, Chevalier P, Millat G (2017) Drosophila and human FHOD family formins nucleate actin filament length in muscle sarcomeres. Dev Cell 31:215–228
Kan-o M, Takeya R, Abe T, Kitajima N, Nishida M, Tominaga R, Kurose JA, Hutchinson KR, Chu M, Nam KH, Valdez JM, Wong PK, Granzier HL, Gregorio CC (2015) Knockout of Lmod2 results in shorter thin filaments followed by dilated cardiomyopathy and juvenile lethality. Proc Natl Acad Sci U S A 112:13573–13578
Patel AA, Oztag Durer ZA, van Loon AP, Bremer KV, Quinlan ME (2018) Drosophila and human FHOD family formin proteins nucleate actin filaments. J Biol Chem 293:532–540
Pappas CT, Mayfield RM, Henderson C, Jamilpour N, Cover C, Hernandez Z, Hutchinson KR, Chu M, Nam KH, Valdez JM, Wong PK, Granzier HL, Gregorio CC (2015) Knockout of Lmod2 results in shorter thin filaments followed by dilated cardiomyopathy and juvenile lethality. Proc Natl Acad Sci U S A 112:13573–13578
Pollard TD, Cooper JA (1986) Actin and actin-binding proteins. A critical evaluation of mechanisms and functions. Annu Rev Biochem 55: 987–1035
Kee AJ, Gunning PW, Hardeman EC (2009) Diverse roles of the actin cytoskeleton in striated muscle. J Muscle Res Cell Motil 30:187–197
Kimura A, Harada H, Park JE, Nishi H, Satoh M, Takahashi M, Hiroi S, Sasaoa T, Ohbuchi N, Nakamura T, Koyanagi T, Hwang TH, Choo JA, Chung KS, Hasegawa A, Nagai R, Okazaki O, Nakamura H, Matsuizaka M, Sakamoto T, Hossia Y, Imaizumi T, Sasazuki T (1997) Mutations in the cardiac troponin I gene associated with hypertrophic cardiomyopathy. Nat Genet 16:379–382
Kooij V, Viswanathan MC, Lee DI, Rainer P, Schmidt W, Kronert WA, Harding SE, Kass DA, Bernstein SI, Van Eyke J, Cammarato A (2016) Profilin modulates sarcomeric organization and mediates cytoskeleton hypertrophy. Cardiovasc Res 110:238–248
Kremenew E, Makkonen MH, Skwarek-Maruszewska A, Gateva G, Michelot A, Dominguez R, Lappalainen P (2014) Cofilin-2 controls actin filament length in muscle sarcomeres. Dev Cell 31:215–226
Lai D, Gao J, Bi X, He H, Shi X, Wen S, Chen Y, Yang Y, Ye Y, Fu G (2017) The Rho kinase inhibitor, fasudil, ameliorates diabetes-induced cardiac dysfunction by improving calcium clearance and actin remodeling. J Mol Med (Berl) 95:155–165
Lange S, Gehmlich K, Lun AS, Blondelle J, Dalton ND, Alvarez EA, Zhang X, Bang ML, Absassi YA, dos Remedios CG, Peterson KL, Chen J, Ehler E (2016) MLP and CARK are linked to chronic PKCalpha signalling in dilated cardiomyopathy. Nat Commun 7:12120
Li D, Hallett MA, Zhu W, Rubart M, Liu Y, Yang Z, Chen H, Hannelin LS, Chan RJ, Schwartz RJ, Field LJ, Atkinson SJ, Shou W (2011) Dishevelled-associated activator of morphogenesis 1 (Daam1) is required for heart morphogenesis. Development 138:303–315
Lin YH, Li J, Swanson ER, Russell B (2013) CapZ and actin capping dynamics increase in myocytes after a bout of exercise and abates in hours after stimulation ends. J Appl Physiol (1985) 114:1603–1609
Lin YH, Warren CM, Li J, McKinsey TA, Russell B (2016) Myofibril growth during cardiac hypertrophy is regulated through dual phosphorylation and acetylation of the actin capping protein CapZ. Cell Signal 28:1015–1024
Pyle WG, La Rotta G, de Tombe PP, Sumandea MP, Solano RJ (2006) Control of cardiac myofilament activation and PKC-betall signaling through the actin capping protein, CapZ. J Mol Cell Cardiol 41:537–543

Randall TS, Ehler E (2014) A formin-g role during development and disease. Eur J Cell Biol 93:205–211

Rangrez AY, Hoppe P, Kuhn C, Zille E, Frank D (2017) MicroRNA miR-301a is a novel cardiac regulator of Cofilin-2. PLoS One 12:e0183901

Ribeiro EA, Pinotsis N, Ghisleni A, Salmazo A, Konarev PV, Kostan J, Rangrez AY, Hoppe P, Kuhn C, Zille E, Frank J, Frey N, Frank D (2017) The genetic basis for cardiomyopathy: from mutation identification to mechanistic paradigms. Cell 104:557–567

Smart N, Bollini S, Dube KN, Vieira JM, Zhou B, Davidson S, Yellon D, Smart N, Riegler J, Turtle CW, Lygate CA, McAndrew DJ, Gehmlich K, Maini NC, Cadot B, Way M, Gomes ER (2017) Myofibril contraction and crosslinking drive nuclear movement to the periphery of skeletal muscle. Nat Cell Biol 19:1189–1207

Seidman JG, Seidman C (2001) The genetic basis for cardiomyopathy: from mutation identification to mechanistic paradigms. Cell 104:557–567

Smart N, Bollini S, Dube KN, Vieira JM, Zhou B, Davidson S, Yellon D, Riegler J, Price AN, Muthurangu V, Lythgoe MF, PWT, Riley PR (2011) De novo cardiomyocytes from within the activated adult heart after injury. Nature 474:640–644

Seidman JG, Seidman CE (2010) FHOD1 is a combined actin filament capping and bundling factor that selectively associates with actin arcs and stress fibers. J Cell Sci 126:1891–1901

Stark K, Esslinger UB, Reinhard W, Petrov G, Winkler T, Komajda M, Isnard R, Charron P, Villard E, Cambien F, Tiren L, Aumont MC, Dubourg O, Trochon JN, Fauchier L, Degroote P, Richter A, Maisch B, Wichter T, Zollbrecht C, Grassl M, Schunklen H, Linsel-Nitschke P, Erdmann J, Baumert J, Illig T, Klopff N, Wichmann HE, Meisinger C, Koenig W, Lichtner P, Meitinger T, Shutlert A, Konig IR, Hetzer R, Heid IM, Regitz-Zagrosek V, Hengstenberg C (2010) Genetic association study identifies HSPB7 as a risk gene for idiopathic dilated cardiomyopathy. PLoS Genet 6:e1001167

Subramanian K, Gianni D, Balla C, Assenza GE, Joshi M, Semigran MJ, Macgillivray TE, Van Eijk JE, Agetti G, Paolocci N, Bamburg JR, Agrawal PB, Del Monte F (2015) Cofilin-2 phosphorylation and sequestration in myocardial aggregates: novel pathogenic mechanisms for idiopathic dilated cardiomyopathy. J Am Coll Cardiol 65:1199–1214

Sussman MA, Welch S, Cambon N, Kleitsky R, Hewett TE, Price R, Witt SA, Kimball TR (1998) Myofibril degeneration caused by tropomodulin overexpression leads to dilated cardiomyopathy in juvenile mice. J Clin Invest 101:51–61

Suurmeijer AJ, Clement S, Francesconi A, Bocchi L, Angelini A, Van Veldhuisen DJ, Spagnoli LG, Gabbiani G, Orlandi A (2003) Alpha-actin isoform distribution in normal and failing human heart: a morphological, morphometric, and biochemical study. J Pathol 199:387–397

Taniguchi K, Takeya R, Suetogu S, Kan OM, Narusawa M, Shiose A, Tominga R, Sumimoto H (2009) The mammalian formin FHod3 regulates actin assembly and sarcomere organization in striated muscles. J Biol Chem 284:29873–29881

Tardiff JC (2011) Thin filament mutations: developing an integrative approach to a complex disorder. Circ Res 108:765–782

Tondeleir D, Vandenau M, Vandeckerkhove J, Ampe C, Lambrecht A (2009) Actin isoform expression patterns during mammalian development and in pathology: insights from mouse models. Cell Motil Cytoskeleton 66:798–815

Tsukada T, Pappas CT, Moroz N, Antin PB, Kostyukova AS, Gregorio CC (2010) Leiomodin-2 is an antagonist of tropomodulin-1 at the pointed end of the thin filaments in cardiac muscle. J Cell Sci 123:3136–3145

Tucker NR, McElanen MA, Hu D, Ye J, Parsons VA, Mills RW, Clausen M, Dolmatova E, Shea MA, Milan DJ, Scott NS, Lindsay M, Lubitz SA, Domian IJ, Stone JR, Lin HEllinor PT (2017) Novel mutation in FLNC (Filamin C) causes familial restrictive cardiomyopathy. Circ Cardiovasc Genet 10:e001780

Ushijima T, Fujimoto N, Matsuuya S, Kan OM, Kiyonari H, Shioi G, Ushijima T, Fujimoto N, Matsuuya S, Kan OM, Kiyonari H, Shioi G, Katae Y, Yamassaki S, Takeya R, Sumimoto H (2018) The actin-organizing formin protein FHod3 is required for postnatal development and functional maintenance of the adult heart in mice. J Biol Chem 293:148–162

Vandeckerkhove J, Weber K (1978) At least six different actins are expressed in a higher mammal: an analysis based on the amino acid sequence of the amino-terminal tryptic peptide. J Mol Biol 126:783–802

Watkins H, McKenna WJ, Thierfelder L, Suk HJ, Aan R, D’Onoghue A, Spriro P, Matsumori A, Moravec CS, Seidman JG, Seidman CE (1995) Mutations in the genes for cardiac troponin T and alpha-tropomyosin in hypertrophic cardiomyopathy. N Engl J Med 332:1058–1064

Wilson AJ, Schoenauer R, Ehler E, Agarkova I, Bennett PM (2014) Cardiomyocyte growth and sarcomerogenesis at the intercalated disc. Cell Mol Life Sci 71:165–181

Wooten EC, Hebl VB, Wolf MJ, Greytak SR, Draper I, Calvino JE, Kapur NK, Marson RS, Kilel J, Ommen SR, Bos JM, Ackerman MJ, Hugins GS (2013) Formin homology 2 domain containing 3 variants associated with hypertrophic cardiomyopathy. Circ Cardiovasc Genet 6:10–18

Wu T, Mu Y, Bogomolovas J, Fang X, Veevers J, Nowak RB, Pappas CT, Gregorio CC, Evans SM, Fowler VM, Chen J (2017) HSPB7 is indispensable for heart development by modulating actin filament assembly. Proc Natl Acad Sci U S A 114:11956–11961

Yang FH, Pyle WG (2012) Reduced cardiac CapZ protein protects hearts against acute ischemia-reperfusion injury and enhances preconditioning. J Mol Cell Cardiol 52:761–772