



HAL
open science

Does the Actin Network Architecture Leverage Myosin-I Functions?

Julien Pernier, Kristine Schauer

► **To cite this version:**

Julien Pernier, Kristine Schauer. Does the Actin Network Architecture Leverage Myosin-I Functions?. Biology, 2022, 11 (7), pp.989. 10.3390/biology11070989 . hal-03867074

HAL Id: hal-03867074

<https://hal.science/hal-03867074>

Submitted on 23 Nov 2022

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

Review

Does the Actin Network Architecture Leverage Myosin-I Functions?

Julien Pernier ¹  and Kristine Schauer ^{2,*} 

¹ Institute for Integrative Biology of the Cell (I2BC), Centre National de la Recherche Scientifique (CNRS), Commissariat à l'Énergie Atomique et aux Énergies Alternatives (CEA), Université Paris-Saclay, 91198 Gif-sur-Yvette, France; julien.pernier@i2bc.paris-saclay.fr

² Tumor Cell Dynamics Unit, Inserm U1279, Gustave Roussy Institute, Université Paris-Saclay, 94800 Villejuif, France

* Correspondence: kristine.schauer@gustaveroussy.fr

Simple Summary: Here, we review the known characteristics and functions of proteins called myosin-I. These mechanoenzymes belong to an ancient family of actin-dependent motors, found throughout eukaryotic cells, which are characterized by intracellular membrane-bound compartments. We elaborate on the surprising fact that many different functions have been attributed to these proteins, and highlight that we now need to understand how their enzymatic activity supports these functions. We propose to focus on the remodeling of the actin cytoskeleton, a higher-order dynamic scaffold typical for eukaryotic cells.

Abstract: The actin cytoskeleton plays crucial roles in cell morphogenesis and functions. The main partners of cortical actin are molecular motors of the myosin superfamily. Although our understanding of myosin functions is heavily based on myosin-II and its ability to dimerize, the largest and most ancient class is represented by myosin-I. Class 1 myosins are monomeric, actin-based motors that regulate a wide spectrum of functions, and whose dysregulation mediates multiple human diseases. We highlight the current challenges in identifying the “pantograph” for myosin-I motors: we need to reveal how conformational changes of myosin-I motors lead to diverse cellular as well as multicellular phenotypes. We review several mechanisms for scaling, and focus on the (re-) emerging function of class 1 myosins to remodel the actin network architecture, a higher-order dynamic scaffold that has potential to leverage molecular myosin-I functions. Undoubtedly, understanding the molecular functions of myosin-I motors will reveal unexpected stories about its big partner, the dynamic actin cytoskeleton.

Keywords: actin; myosin-I; monomeric motor; mechanical transduction; membrane tension; cholesterol transport; autophagosome–lysosome fusion; glucose metabolism; chirality; exocytosis



Citation: Pernier, J.; Schauer, K. Does the Actin Network Architecture Leverage Myosin-I Functions? *Biology* **2022**, *11*, 989. <https://doi.org/10.3390/biology11070989>

Academic Editors: Tina L. Tootle, Margot E. Quinlan and William J. Lehman

Received: 19 April 2022

Accepted: 26 June 2022

Published: 29 June 2022

Publisher's Note: MDPI stays neutral with regard to jurisdictional claims in published maps and institutional affiliations.



Copyright: © 2022 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<https://creativecommons.org/licenses/by/4.0/>).

1. Introduction

The actin cytoskeleton plays a crucial role in cell morphogenesis and function. The main partners of cortical actin are the molecular motors of the myosin superfamily. Myosins are mechanoenzymes that use the hydrolysis of ATP as an energy source to interact with actin filaments, and to generate force on them. Myosins are present in almost all eukaryotes, missing only in the metamonads *Trichomonas vaginalis* and *Giardia lamblia*, and some red algae, *Cyanidioschyzon merolae* and *Chondrus crispus* [1]. Phylogenetic analyses of the myosin gene family indicate that 2–6 myosin genes, encoding different protein domain architectures, were present in the last eukaryotic common ancestor (LECA), revealing an ancestral role of myosins in eukaryotic evolution [1]. It has been proposed that there are at least 79 distinct myosin classes in extant eukaryotes, 13 of which are found in humans [2]. Indeed, humans also have one of the highest numbers of myosin genes (total of 38) and one of the richest

protein domain diversities attached to the myosin motor domain [1]. Yet, the function of most of these myosins is not known and an understanding of myosin diversity is lacking.

Myosin research started with muscle myosin, a class 2 myosin. The mechano-chemical characteristics of the muscle myosin heavy chain has strongly influenced our thinking about these actin-dependent motor proteins. For instance, historic classification groups myosins into two categories, named conventional myosin-II, with their long tails necessary for dimerization, and unconventional myosins, with distinct tails associated with different roles. Class 2 myosins (Myh genes in humans) dimerize and slide antiparallel actin filaments against each other. In muscle, these activities lead to muscle contraction, whereas in other cells, non-muscle myosin-II a/b (Myh9 and Myh10 in humans) crosslinks actin to (i) produce the actin retrograde flow [3], (ii) regulate cell cortex tension [4–6], or (iii) pull on adhesive structures [7]. In addition to myosin-II, some other myosins, such as myosin-V (Myo5) and -XVIII (Myo18), act as dimers that interact through their C-terminal coiled-coils. However, the majority of myosins are considerably divergent, including myosins I, III, VI, VII, IX, X, XV, and XIX (Myo genes in human), and act as monomers [8].

To date, only amorphoan, apicomplexan and plant myosins have been studied in detail, and functional data are missing for most of the myosin classes. Although our understanding of myosin functions is heavily based on myosin-II and its ability to dimerize, the largest and most ancient class is represented by monomeric myosin-I. In this review, we will focus on these important myosins. We will highlight current challenges to understand how a single-headed, actin-based motor could regulate diverse functions, and how its dysregulation mediates multiple human diseases. We review several mechanisms that could leverage the molecular conformational changes of myosin-I through scales, and thus function as a cellular or organism-level “pantograph” for myosin-I motors.

1.1. Molecular Domains of Myosin-I

Class 1 myosins are non-processive monomeric motors that are composed of three main domains: head, neck and tail [9]. It has been proposed to classify myosin-I members into five subclasses based on a phylogenetic analysis of their motor and additional domain structure [1]. Four of these subclasses are present in humans, and these orthologs have been used to name the subclasses of myosin-I, namely subclasses c/h, d/g, and a/b as well as less related f isoforms representing long tails (Figure 1). Additionally, a myosin-Ik subclass was found that was lost in metazoans and is only present in choanoflagellates, filastereans, ichthyosporeans, and *Thecamonas trahens* [1].

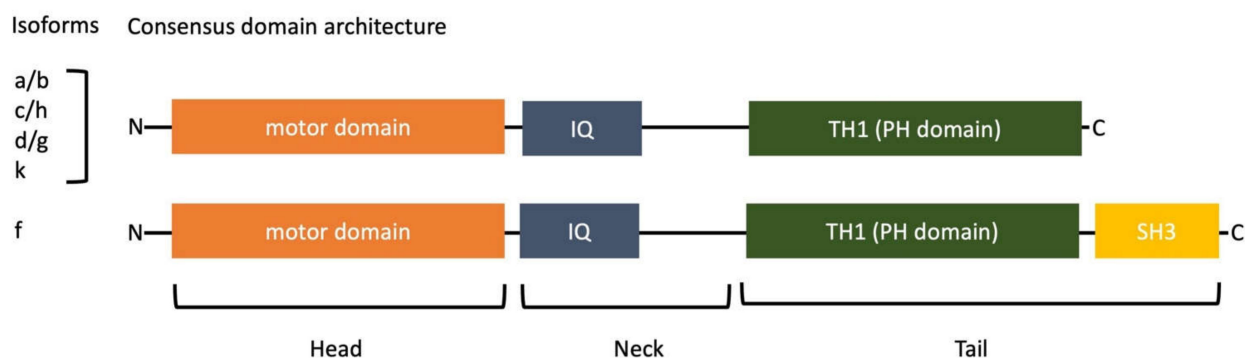


Figure 1. Characteristic protein domain organization of class 1 myosins modified from [1].

The N-terminus of all class 1 myosins is called the head domain and contains the catalytic motor domain. It binds filamentous actin (F-actin) in an ATP-regulated manner (Figure 2). ATP-dependent motor activity leads to a movement of myosin-I towards the plus end of actin filaments (barbed ends). This conserved catalytic domain is followed by a neck region that mostly has one or more consensus sequences called the IQ motif. They can bind regulatory proteins, such as calmodulin or calmodulin-like light chains. Finally, the

C-terminal tail contains a myosin-I family tail homology 1 (TH1) domain, which includes a pleckstrin homology (PH) domain known to bind a variety of anionic phospholipids. Subclasses c/h, d/g, a/b and k consist of a myosin head domain followed by a neck, composed of IQ repeats (PF00612), and a C-terminal myosin TH1 domain (PF06017). Long-tailed isoforms of myosin-If (Myo1E and Myo1F) have a myosin head domain followed by one IQ motif, a myosin TH1 domain and a C-terminal proline rich domain, which includes a src-homology (SH3) domain, involved in the binding of other proteins. The characteristic protein domain organization results in the localization of myosin-I to cellular membranes, the plasma membrane and intracellular organelles. Phosphoinositides, which are dynamic membrane phospholipids and master regulators in intracellular trafficking and signaling, control the specific binding of myosin-I to cellular membranes through the PH domain [10]. Moreover, the preference of motor domains for different actin filament populations could influence cellular localization [11]. Finally, the presence of additional domains (such as the SH3 domain) determines specific interactions with other protein-binding partners. Collectively, class 1 myosins connect the actin cytoskeleton to cellular membranes rather than crosslinking actin, which is the main function of class 2 myosin isoforms.

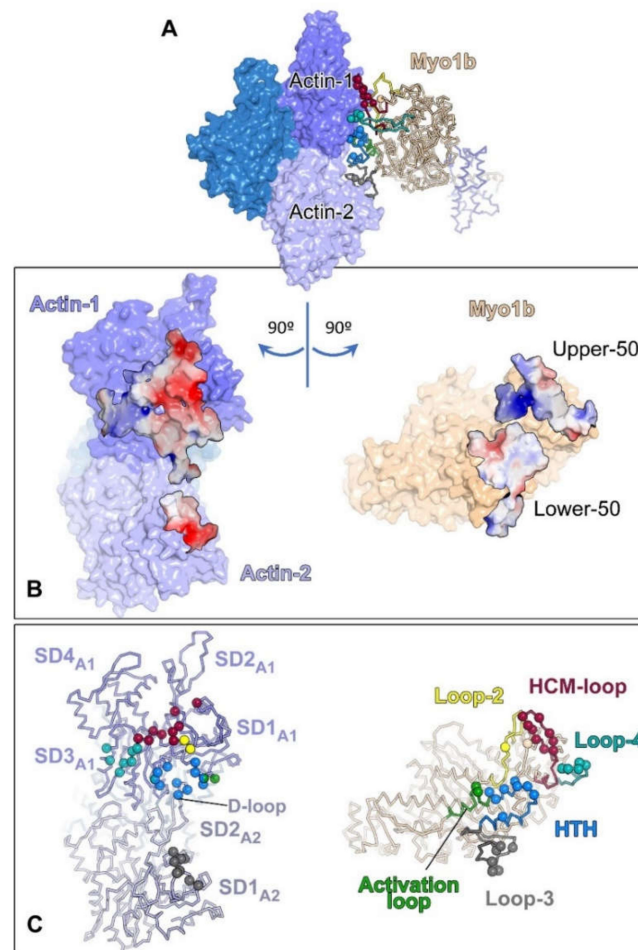


Figure 2. The actin–myosin interface. (A) The Myo1b motor domain bound to F-actin (with only 3 actin subunits). (B) Actin (left) and myosin (right) surfaces (within a 5 Å distance of each other) are colored according to the surface electrostatic potential distribution (red −5; blue 5 K_BT/ec). (C) Residues making inter-protein contacts (within 4 Å distance) are shown as balls. Right, the myosin actin-binding structural elements are shown in different colors. Left, actin residues are colored depending on the myosin structural elements that contact them. This figure was provided by Olena Pylypenko, UMR144/Institut Curie. The figure was made based on an experimental cryo-EM structure of Myo1B (PDB ID 6C1H) from (Mentes et al. [12]).

1.2. Myosin-I Members Have Diverse Functions

Myosin-I motors represent the largest and the most ancient myosin class in eukaryotes. The number of myosin-I genes strongly varies in different organisms, e.g., the yeast *Saccharomyces cerevisiae* has two long-tailed myosins, Myo3p and Myo5p [13], whereas *Schizosaccharomyces pombe* has only Myo1p [14]. *Dictyostelium* has seven myosin-I isoforms (Myo1A, B, C, D, E, F and K), three short-tailed (1A, 1E and 1F), three long-tailed (1B, 1C and 1D) and another myosin-I (Myo1K) with no tail [15]. Eight members (Myo1A, B, C, D, E, F and H) are known in humans. Given the ancient nature of myosin-I, it is not surprising that very diverse functions have been associated with this motor protein in different organisms and tissues [9]. The non-exhaustive functions of the different human subtypes are listed below and are summarized in Table 1.

Table 1. Localizations and functions of myosin 1 paralogs.

Myosin Name	Localizations	Functions	References
Myo1A	Intestine	Linker between actin bundles and the microvillus membrane in mice	Benesh, A.E. [16]
		Interacts with EPHB2 and controls cell repulsion and morphology (Hek293T or HCT116 cells)	Prospéri, M.-T. [17] Prospéri, M.-T. [18]
Myo1B	Lung, liver, heart, and brain	Regulates the formation of filopodia in growth cones in neurons	Iuliano, O. [19]
		Promotes the formation of tubules at the Trans-Golgi Network (HeLa cells)	Almeida, C.G. [20]
		Facilitates tube extraction (in vitro, rat Myo1B)	Yamada, A. [21]
		Acts as an actin depolymerase (in vitro, rat Myo1B)	Pernier, J. [22]
		Affects branched F-actin (in vitro, rat Myo1B)	Almeida, C.G. [20] Pernier, J. [23]
Myo1C	Ubiquitously expressed	Mechanical transduction in the ear (mouse Myo1c)	Lin, T. [24]
		Regulation of cellular membrane tension (<i>Dictyostelium</i>)	Dai, J. [25]
		Cholesterol transport (HeLa cells)	Brandstaetter, H. [26]
		Tethering of vesicles (mouse Myo1c, hTERT-RPE1 cells)	Boguslavsky, S. [27] Capmany, A. [28]
		Autophagosome-lysosome fusion (HeLa cells)	Brandstaetter, H. [29]
		Chirality establishment in <i>Drosophila</i>	Lebreton, G. [30] Pyrpassopoulos, S. [31]
		Docking of GLUT4-containing vesicles to the plasma membrane in muscle and adipocytes	Boguslavsky, S. [27] Bose, A. [32] Bose, A. [33] Yip, M.F. [34] Chen, X.-W. [35] Huang, S. [36]
Controls exocytosis in secretory cells	Tiwari, A. [37] Kittelberger, N. [38]		
Myo1D	Widely expressed, high expression in brain	Chromatin modifications to gene transcription and cell cycle progression in nucleus (HeLa cells)	Sarshad, A. [39]
		Influences the establishment of rotational planar cell polarity in epithelial cells of the trachea	Hegan, P.S. [40]
Myo1E	Hematopoietic cells	Involved in clathrin-mediated endocytosis	Lechler, T. [41] Cheng, J. [42]
Myo1F	Hematopoietic cells	Regulates adhesion	Kim, S.V. [15]
Myo1G	Hematopoietic cells	Involved in phagocytosis and exocytosis processes	Maravillas-Montero, J.L. [43]

Moreover, mutations in myosin-I motors have been found in several diseases [9], including kidney disease [44–48], deafness [49–53], arteriosclerosis [54] and have been linked to the mandibular prognathism phenotype [55] (summarized in Table 2).

Table 2. Diseases associated with myosin 1 paralog mutations.

Myosin Name	Amino Acid Mutation	Effect and Associated Disease	Reference
Myo1A	Insertion between 349 and 350	Deafness	Donaudy, F. [51]
	V306M	Deafness	
	E385D	Affect ATPase activity Deafness	
	G662E	Deafness	
	G647D	Deafness	
	S797F	Deafness	
	S910P	Deafness	
Myo1C	690STOP	No recruitment of Neph1, glomerular disease	Arif, E. [44]
	Y61G	Increased sensitivity to inhibition, hair cells defect	Holt, J.R. [50]
	R156W	Decreased myosin duty ratio and force sensitivity, hearing loss	Lin, T. [24]
	K111A (in Mouse)	Glucose uptake defects	Toyoda, T. [56]
Myo1E	A159P	Abnormal localization and function, glomerulosclerosis	Mele, C. [46]
	Y695STOP	Loss of calmodulin binding at the tail domain of Myo1E, glomerulosclerosis	
	A159P	No efficient assembly of actin cables along cell-cell junctions, glomerular disease	Bi, J. [48]
Myo1F	I502V	Destabilized actin binding site and ATP binding site, hearing loss	Baek, J.-I. [53]
Myo1H	P1001L	Mandibular prognathism	Sun, R. [57]

Finally, several knock out murine models of myosin-I paralogs have been established and their phenotypes are summarized in Table 3.

Table 3. Genetic studies and experiments using murine models of myosin-I.

Myosin Name	Genetic Modification	Phenotypes	References
Myo1A	Complete knockout	No overt phenotypes at the whole animal, defects in microvillar membrane morphology and in brush-border organization	Tyska, M.J. [58]
Myo1B	-	-	
Myo1C	Complete knockout	Retinal phenotypes only	Arif, E. [59]
Myo1C	Podocytes-specific knockout	Downregulation of canonical and non-canonical TGF- β pathways	Arif, E. [59]
Myo1D	Complete knockout	Perturbed planar cell polarity of epithelial cells of the trachea, change of velocity and linearity of cilia-driven movement, loss of asymmetric clustering of cilia of ependymal cells and left-right positioning of the clusters, no obvious motor defects of rats, no obvious differences in kidney and liver morphology	Hegan, P.S. [40]

Table 3. Cont.

Myosin Name	Genetic Modification	Phenotypes	References
Myo1E	Complete knockout	Nephrotic syndrome and focal segmental glomerulosclerosis	Krendel, M. [45]
Myo1E	Podocytes-specific knockout	Proteinuria, podocyte foot process effacement, glomerular basement membrane disorganization, anormal glomerular filtration	Chase, S.E. [60]
Myo1F	Complete knockout	Increased susceptibility to infection by <i>Listeria monocytogenes</i> and an impaired neutrophil response	Kim, S.V. [61]
Myo1G	Complete knockout	Abnormalities in the adhesion ability and chemokine-induced directed migration in B lymphocytes	Maravillas-Montero, J.L. [43]
Myo1H	Complete knockout	Severe cyanosis and death within the first four postnatal hours	Spielmann, M. [62]

The myosin a/b subclass, Myo1A, is expressed in intestine [16] where it acts as a linker between actin bundles and the microvillus membrane. Myo1B has three different isoforms (Myo1Ba, b and c) containing four to six IQ motifs. It is expressed in many tissues such as lung, liver, heart, and brain [63]. Myo1B interacts with the transmembrane tyrosine kinase receptors EPHB2 and controls cell repulsion and morphology [17,18]. Myo1B also regulates the formation of filopodia in growth cones and controls the extension of branched actin networks that impact axon elongation [19]. Moreover, Myo1B has been shown to have intracellular functions, promoting the formation of tubules at the Trans-Golgi-Network (TGN) [20]. It has been proposed that its mechano-enzymatic activity bends membranes and facilitates tube extraction [21].

The myosin c/h subclass is exemplary of myosin-I regulation of a broad range of biological processes and is the ubiquitously expressed Myo1C isoform. Myo1C has been implicated in a variety of different functions, including mechanical transduction in the ear [24], the regulation of cellular membrane tension [25], cholesterol transport [26], tethering of vesicles [27,28], autophagosome–lysosome fusion [29], the regulation of metabolism [27,32–36], and the establishment of chirality in *Drosophila* [30]. Interestingly, mutations in Myo1C manifest in different diseases (Table 2), from glucose-uptake defects [56] to hearing loss [24]. The best-studied function of Myo1C is its role in the docking of GLUT4-containing vesicles to the plasma membrane in response to insulin stimulation [27,32–36]. More generally, Myo1C has been shown to control exocytosis in secretory cells and has been implicated in the VEGF-induced delivery of VEGFR2 to the cell plasma membrane [37] and in surfactant exocytosis (lipo-protein secretion) [38]. Concomitantly, although Myo1C shows a strong accumulation at the plasma membrane in cholesterol-rich lipid microdomains [26], it can also be found in minor pools such as on recycling endosomes [26] or at the proximity of the Golgi complex [28]. Myo1C's paralog, Myo1H, although little studied, has recently been implicated in Mandibular Prognathism [14].

The myosin d/g subclass, Myo1D, is widely expressed, with the highest expression being seen in the brain. Myo1D has two IQ motifs that bind calmodulin [64]. Ca²⁺ binding to calmodulin can inhibit ATPase activity [65]. In rats, Myo1D functions at adherens junctions. It also influences the establishment and/or maintenance of rotational planar cell polarity in the ciliated tracheal and ependymal epithelial cells [40]. Myo1G is specifically expressed in hematopoietic cells [66]. In B lymphocytes, it is involved in the processes of phagocytosis and exocytosis [43].

The long-tailed myosin e/f subclass are both long-tailed isoforms expressed in hematopoietic cells. The widely expressed long-tailed Myo1E and its homologs in *S. cerevisiae* (Myo3p and Myo5p) interact through their SH3 domain with the Wiskott–Aldrich syndrome proteins (WASP). This interaction promotes Arp2/3-mediated actin polymerization during clathrin-mediated endocytosis [41,42]. Moreover, Myo1E localizes to actin

polymerization sites in lamellipodia [67]. Myo1F is a long-tailed myosin that, in addition to a TH1, also contains a C-terminal proline-rich domain and a src-homology (SH3) domain, involved in the binding of other proteins. It is selectively expressed in neutrophils and regulates adhesion to the extracellular environment [61].

Collectively, and as reviewed in more detail previously [9], a wide range of biological processes and complex cellular phenomena have been shown to depend on individual myosin-I isoforms in different cellular and developmental systems. In addition to an enzymatic motor domain, most myosin-I isoforms contain a very short tail, only consisting of a pleckstrin homology (PH) domain for phosphatidylinositol binding [68]. Therefore, it is unclear how these relatively simple molecules and their mechano-enzymatic activity control a wide spectrum of cellular functions.

2. Challenges to Leverage the Molecular Conformational Changes of Myosin-I through Scales

It is intriguing how many different cellular roles, related to membrane dynamics and trafficking, are played by individual myosin-I motors [9]. The challenge holds up to connect the mechano-enzymatic activity of these molecules to cellular and organism-level functions [9], including impressive phenotypes of chirality regulation in *Drosophila*. In other words, it is high time we understood the scaling from molecular conformational changes of a rather simple molecule to cell and organism functions, and to identify the “pantograph” for myosin-I motors. We particularly argue that we need to leave behind our thinking of myosin functions based on dimerization (as found for myosin-II) or attributed to the tail domain only, and consider how the conformational changes of the motor domain of myosin-I could support different functions in a monomeric form. We review several mechanisms that have been proposed for scaling, from the deformation of membranes to the regulation of membranes and cortical tension, and finally focus on the emerging role of class 1 myosins in the remodeling of a higher-order dynamic scaffold, represented by the actin cytoskeleton.

2.1. Membrane Remodeling

Given that myosin-I isoforms link membranes to the actin cytoskeleton, it has been proposed that they also deform membranes and participate in tubulation at the plasma membrane or the surface of organelles [9]. This function is often associated with plasma membrane invagination during different types of endocytosis [69,70], changes in plasma membrane deformability (or plasma membrane tension) during protrusion formation [71–74] or intracellular membrane trafficking [20,75–77]. Membrane deformation functions of myosin-I have also been shown to occur working with processive microtubule-based motors of the kinesin family [20,78]. Strong experimental support for myosin-I function in membrane deformation comes from in vitro studies. It has been shown that the myosin-I member Myo1B can tubulate giant unilamellar vesicles along fascin-bundled actin in an in vitro reconstitution assay [21]. However, it is surprising that the PH-domain-dependent membrane attachment and dynamic actin binding provides stable anchorages for membrane deformations. Although the regulation of membrane tubulation, and therefore the control of intracellular trafficking could impact many functions indirectly, some of the phenotypes associated with myosin-I depletion/mutations are difficult to explain. For instance, chirality phenotypes rely on asymmetric cellular activity. Thus, membrane remodeling only provides limited explanations of how molecular myosin-I functions could scale over several dimensions.

2.2. Membrane and Cortical Tension

The actin-membrane linker function of myosin-I could also regulate cortical tension, a mechanical property of the plasma membrane with the underlying actin cortex [79]. Yet, cortical tension results from applied contractile stresses that pull on actin filaments through motor crosslinking, for instance. It has been proposed that cortical tension is most

prominently controlled by myosin-II activity [4–6]. Because monomeric myosin-I motors do not crosslink actin, it is less obvious how they could develop contractile stresses. The molecular role of myosin-I at the cell cortex is not well understood. Interestingly, cortex tension strongly cross-talks with cell–substrate and cell–cell adhesion in a multicellular context. Indeed, myosin-I motors have been localized at cell–cell adhesions and adherens junctions and have also been implicated in ephrin receptor B2 (EphB2)-dependent cell–cell repulsion [17]. The balance between cell–cell adhesion and cell–cell repulsion is important for tissue patterning and homeostasis, and thus could regulate chirality establishment or the maintenance of rotational planar cell polarity [40]. Yet, the maintenance of membrane and/or cortical tension, as well as cell–cell adhesion functions, rely on the underlying actin cytoskeleton and therefore the role of myosin-I in the control of actin architecture needs to be further considered.

2.3. Role in Actin Remodeling

A mechanism that could allow one to scale the mechano-enzymatic activity of myosin-I over several dimensions, from cellular compartments to whole cells and organisms, is the dynamic remodeling of a bigger scaffold, the actin cytoskeleton. Indeed, several lines of evidence exist that myosin-I changes actin architecture in vitro, in cellulo and in vivo [22,23,43,58,80]. For instance, several myosin-I motors have been proposed to regulate cortical actin architecture in the neuronal growth cone [80] and in B cells at the immunological synapse [81]. Experiments on adhesive micropatterns, which provide good control and reproducibility to monitor the actin cytoskeleton [82,83], confirm that the depletion of Myo1C leads to the loss of cellular F-actin [28]. Strong evidence for a direct role in actin reorganization of class 1 myosins comes from biochemical studies of human Myo1B which has been found to act as an actin de-polymerase [22].

Particularly, it is emerging that myosin-I motors regulate the balance between actin bundles and Arp2/3-dependent branched actin. In vitro and in vivo data suggest that myosin-I motors avoid tropomyosin-coated actin filaments [84–87] and instead prefer Arp2/3-nucleated actin [88]. Moreover, the depletion of Myo1C mimics the loss of the Arp2/3 complex, whereas the manipulation of Myo1B affects the distribution of Arp2/3 and associated branched F-actin [20], and flattens branched actin filaments in vitro [23]. Indeed, it has been highlighted that many class 1 myosins appear to be able to recruit, directly or indirectly, the Arp2/3 complex [89]. In *Dictyostelium*, Myo1E and Myo1F are involved in actin dynamics at the leading edge, by interacting with Arp2/3 through CARMIL (capping protein, Arp2/3, myosin-I linker protein) [90]. The long-tailed class 1 myosins, Myo1E in human, and Myo3p and Myo5p in yeast, also interact with the actin-polymerization factors neural Wiskott–Aldrich syndrome protein (N-WASP) and with the Arp2/3 complex, through a sequence at the end of their long tail domain [41,91]. Yet, although yeast class 1 myosins facilitate Arp2/3 nucleation through their tail domain, it has been recently suggested that actin network growth relied on their motor domain [92]. Because a deficiency of myosin motors could not be rescued by increasing actin nucleation, it was proposed that class 1 myosins stimulate actin filament elongation. It was suggested that the myosin-I motor domain facilitates the addition of new actin monomers to existing filaments, rather than supporting nucleation [92]. Thus, in addition to indirectly affecting actin dynamics by recruiting factors that are involved in actin nucleation, polymerization or stabilization, myosin-I could regulate actin dynamics directly through its motor domain. Interestingly, *Drosophila* Myo1C and Myo1D control cellular and larvae chirality [30,31], which were thought to rely on the gliding of actin filaments in circular paths revealed in in vitro assays. Alternatively, the higher-scale propagation of the chiral Arp2/3 molecule by the actin network could be envisioned. Thus, we propose to revisit our understanding of class 1 myosins as motors involved in actin rearrangements [22,23,28,92].

3. Conclusions

Actin network organization is at the center of mammalian cell morphogenesis and function. Thus, to fully understand this relationship, we must understand the role and transitions between different types of actin networks. Because myosin motor proteins are prime partners of F-actin, it is important to obtain a true, molecular comprehension of the function of the ancient and ubiquitously expressed class I myosins. A wide range of biological processes have been shown to depend on them in different cellular and developmental systems. However, the molecular mechanisms of different functions achieved by this single-headed, non-dimerizing motor is not clear. After revealing different processes relying on myosin-I, it is now important to integrate the cell biological, biochemical, biophysical and structural features of myosin-I motors.

An emerging, or re-emerging, function of class 1 myosins is the regulation of the actin architecture, which has the potential to leverage molecular myosin-I functions to diverse cellular as well as multicellular phenotypes. In this context, we particularly need to re-evaluate the function of the catalytic motor domain in actin reorganization. Could the motor domain regulate actin dynamics and rearrangements? Could the interaction of the myosin-I motor domain with actin impact the global actin architecture? For this, it is essential to piece together different approaches in continued interdisciplinary research across scales, from molecules and networks to cells and organisms. A strong impact could have in vitro bottom-up approaches, which not only test biochemical activity, but also build up in size and include geometrical constraints mimicking cell constituents, as well as cell dimensions. For instance, assays on fluid-supported lipid bilayers, which mimic cell membranes [22], are important to better estimate the impact of force generation by myosin-I motors. Such in vitro reconstitution approaches need to be further integrated with scalable micropatterned environments, which increase dimensions and allow the study of emerging properties, such as the network architecture of the actin cytoskeleton. Together, such in vitro systems will allow further testing into how the actin network organization could be controlled by non-conventional myosins. Further, their comparison with cells could reveal novel mechanistic explanations for the pleiotropic roles played by these motors in cells and at the scale of organisms. This will provide a foundation to understand functions such as mechano-electrical transduction in the ear [24], the regulation of cellular membrane tension [25], cholesterol transport [26], tethering secretory vesicles for glucose uptake [27,28], autophagosome–lysosome fusion [29], chirality establishment in *Drosophila* [30] and potentially unexpected novel ‘moon light functions’ of Myo1C in the nucleus [39]. Understanding the molecular function of myosin-I motors will also facilitate the development of targeted therapeutics. Only a few inhibitors that target the conserved motor domain of class 1 myosins have been described, for instance, the natural compound pentachloropseudilin (PCIP), a reversible and allosteric inhibitor of myosin ATPase activity. Myosin-I inhibitors are employed as highly species-specific fungicide [93]. Selective myosin-I inhibitors and activators could have therapeutic potential for the treatment of myosin dysfunctions or overactivity, or could be used to specifically target pathogenic eukaryotes. Finally, relating myosin-I activity to the organization of the actin network will shed light on the exciting story about the extensive remodeling of this cytoskeleton system across eukaryotes.

Author Contributions: Writing—review and editing, K.S. and J.P.; funding acquisition, K.S. All authors have read and agreed to the published version of the manuscript.

Funding: This research and the APC was funded by ANR grant MYOCORTEX n° ANR-21-CE13-0010.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Acknowledgments: We would like to acknowledge Olena Pylypenko, Institut Curie, France, for providing Figure 2. We thank all members of the Schauer and Le Clainche team for critical discussions.

Conflicts of Interest: The sponsors had no role in the design, execution, interpretation, or writing of the study.

References

1. Sebé-Pedrós, A.; Grau-Bové, X.; Richards, T.A.; Ruiz-Trillo, I. Evolution and classification of myosins, a paneukaryotic whole-genome approach. *Genome Biol. Evol.* **2014**, *6*, 290–305. [[CrossRef](#)] [[PubMed](#)]
2. Kollmar, M.; Mühlhausen, S. Myosin repertoire expansion coincides with eukaryotic diversification in the Mesoproterozoic era. *BMC Evol. Biol.* **2017**, *17*, 211. [[CrossRef](#)] [[PubMed](#)]
3. Burnette, D.T.; Manley, S.; Sengupta, P.; Sougrat, R.; Davidson, M.W.; Kachar, B.; Lippincott-Schwartz, J. A role for actin arcs in the leading-edge advance of migrating cells. *Nat. Cell Biol.* **2011**, *13*, 371–382. [[CrossRef](#)] [[PubMed](#)]
4. Maître, J.-L.; Niwayama, R.; Turlier, H.; Nédélec, F.; Hiiragi, T. Pulsatile cell-autonomous contractility drives compaction in the mouse embryo. *Nat. Cell Biol.* **2015**, *17*, 849–855. [[CrossRef](#)]
5. Maître, J.-L.; Turlier, H.; Illukkumbura, R.; Eismann, B.; Niwayama, R.; Nédélec, F.; Hiiragi, T. Asymmetric division of contractile domains couples cell positioning and fate specification. *Nature* **2016**, *536*, 344–348. [[CrossRef](#)]
6. Schliffka, M.F.; Tortorelli, A.F.; Özgüç, Ö.; de Plater, L.; Polzer, O.; Pelzer, D.; Maître, J.-L. Multiscale analysis of single and double maternal-zygotic Myh9 and Myh10 mutants during mouse preimplantation development. *eLife* **2021**, *10*, e68536. [[CrossRef](#)]
7. Vigouroux, C.; Henriot, V.; Le Clainche, C. Talin dissociates from RIAM and associates to vinculin sequentially in response to the actomyosin force. *Nat. Commun.* **2020**, *11*, 3116. [[CrossRef](#)]
8. Peckham, M. Coiled coils and SAH domains in cytoskeletal molecular motors. *Biochem. Soc. Trans.* **2011**, *39*, 1142–1148. [[CrossRef](#)]
9. McIntosh, B.; Ostap, E.M. Myosin-I molecular motors at a glance. *J. Cell Sci.* **2016**, *129*, 2689–2695. [[CrossRef](#)]
10. McConnell, R.E.; Tyska, M.J. Leveraging the membrane—Cytoskeleton interface with myosin-1. *Trends Cell Biol.* **2010**, *20*, 418–426. [[CrossRef](#)]
11. Ruppert, C.; Godel, J.; Muller, R.; Kroschewski, R.; Reinhard, J.; Bahler, M. Localization of the rat myosin I molecules myr 1 and myr 2 and in vivo targeting of their tail domains. *J. Cell Sci.* **1995**, *108*, 3775–3786. [[CrossRef](#)]
12. Menten, A.; Huehn, A.; Liu, X.; Zwolak, A.; Dominguez, R.; Shuman, H.; Ostap, E.M.; Sindelar, C.V. High-resolution cryo-EM structures of actin-bound myosin states reveal the mechanism of myosin force sensing. *Proc. Natl. Acad. Sci. USA* **2018**, *115*, 1292–1297. [[CrossRef](#)]
13. Brown, S. Myosins in yeast. *Curr. Opin. Cell Biol.* **1997**, *9*, 44–48. [[CrossRef](#)]
14. Berg, J.S.; Powell, B.C.; Cheney, R.E. A millennial myosin census. *Mol. Biol. Cell* **2001**, *12*, 780–794. [[CrossRef](#)]
15. Kim, S.V.; Flavell, R.A. Myosin I: From yeast to human. *Cell. Mol. Life Sci.* **2008**, *65*, 2128–2137. [[CrossRef](#)]
16. Benesh, A.E.; Nambiar, R.; McConnell, R.E.; Mao, S.; Tabb, D.L.; Tyska, M.J. Differential localization and dynamics of class I myosins in the enterocyte microvillus. *Mol. Biol. Cell* **2010**, *21*, 970–978. [[CrossRef](#)]
17. Prospéri, M.-T.; Lépine, P.; Dingli, F.; Paul-Gilloteaux, P.; Martin, R.; Loew, D.; Knölker, H.-J.; Coudrier, E. Myosin 1b functions as an effector of EphB signaling to control cell repulsion. *J. Cell Biol.* **2015**, *210*, 347–361. [[CrossRef](#)]
18. Prospéri, M.-T.; Pernier, J.; Lachuer, H.; Coudrier, E. Plekh1, a partner of myosin 1 and an effector of EphB2 controls the cortical actin network for cell repulsion. *J. Cell Sci.* **2021**, *134*, jcs258802. [[CrossRef](#)]
19. Iuliano, O.; Yoshimura, A.; Prospéri, M.-T.; Martin, R.; Knölker, H.-J.; Coudrier, E. Myosin 1b promotes axon formation by regulating actin wave propagation and growth cone dynamics. *J. Cell Biol.* **2018**, *217*, 2033–2046. [[CrossRef](#)]
20. Almeida, C.G.; Yamada, A.; Tenza, D.; Louvard, D.; Raposo, G.; Coudrier, E. Myosin 1b promotes the formation of post-Golgi carriers by regulating actin assembly and membrane remodelling at the trans-Golgi network. *Nat. Cell Biol.* **2011**, *13*, 779–789. [[CrossRef](#)]
21. Yamada, A.; Mamane, A.; Lee-Tin-Wah, J.; Di Cicco, A.; Prévost, C.; Levy, D.; Joanny, J.-F.; Coudrier, E.; Bassereau, P. Catch-bond behaviour facilitates membrane tubulation by non-processive myosin 1b. *Nat. Commun.* **2014**, *5*, 3624. [[CrossRef](#)]
22. Pernier, J.; Kusters, R.; Bousquet, H.; Lagny, T.; Morchain, A.; Joanny, J.-F.; Bassereau, P.; Coudrier, E. Myosin 1b is an actin depolymerase. *Nat. Commun.* **2019**, *10*, 5200. [[CrossRef](#)]
23. Pernier, J.; Morchain, A.; Caorsi, V.; Bertin, A.; Bousquet, H.; Bassereau, P.; Coudrier, E. Myosin 1b flattens and prunes branched actin filaments. *J. Cell Sci.* **2020**, *133*, jcs247403. [[CrossRef](#)] [[PubMed](#)]
24. Lin, T.; Greenberg, M.J.; Moore, J.R.; Ostap, E.M. A hearing loss-associated myo1c mutation (R156W) decreases the myosin duty ratio and force sensitivity. *Biochemistry* **2011**, *50*, 1831–1838. [[CrossRef](#)]
25. Dai, J.; Ting-Beall, H.P.; Hochmuth, R.M.; Sheetz, M.P.; Titus, M.A. Myosin I contributes to the generation of resting cortical tension. *Biophys. J.* **1999**, *77*, 1168–1176. [[CrossRef](#)]
26. Brandstaetter, H.; Kendrick-Jones, J.; Buss, F. Molecular roles of Myo1c function in lipid raft exocytosis. *Commun. Integr. Biol.* **2012**, *5*, 508–510. [[CrossRef](#)]
27. Boguslavsky, S.; Chiu, T.; Foley, K.P.; Osorio-Fuentealba, C.; Antonescu, C.N.; Bayer, K.U.; Bilan, P.J.; Klip, A. Myo1c binding to submembrane actin mediates insulin-induced tethering of GLUT4 vesicles. *Mol. Biol. Cell* **2012**, *23*, 4065–4078. [[CrossRef](#)]
28. Capmany, A.; Yoshimura, A.; Kerdous, R.; Caorsi, V.; Lescure, A.; Del Nery, E.; Coudrier, E.; Goud, B.; Schauer, K. MYO1C stabilizes actin and facilitates arrival of transport carriers at the Golgi apparatus. *J. Cell Sci.* **2019**, *132*, jcs225029. [[CrossRef](#)]
29. Brandstaetter, H.; Kishi-Itakura, C.; A Tumbarello, D.; Manstein, D.J.; Buss, F. Loss of functional MYO1C/myosin 1c, a motor protein involved in lipid raft trafficking, disrupts autophagosome-lysosome fusion. *Autophagy* **2014**, *10*, 2310–2323. [[CrossRef](#)]

30. Lebreton, G.; Géminard, C.; Lapraz, F.; Pyrpassopoulos, S.; Cerezo, D.; Spéder, P.; Ostap, E.M.; Noselli, S. Molecular to organismal chirality is induced by the conserved myosin 1D. *Science* **2018**, *362*, 949–952. [[CrossRef](#)]
31. Pyrpassopoulos, S.; Feeser, E.A.; Mazerik, J.N.; Tyska, M.J.; Ostap, E.M. Membrane-bound Myo1c powers asymmetric motility of actin filaments. *Curr. Biol.* **2012**, *22*, 1688–1692. [[CrossRef](#)] [[PubMed](#)]
32. Bose, A.; Guilherme, A.; Robida, S.I.; Nicoloso, S.M.C.; Zhou, Q.L.; Jiang, Z.Y.; Pomerleau, D.P.; Czech, M.P. Glucose transporter recycling in response to insulin is facilitated by myosin Myo1c. *Nature* **2002**, *420*, 821–824. [[CrossRef](#)] [[PubMed](#)]
33. Bose, A.; Robida, S.; Furcinitti, P.S.; Chawla, A.; Fogarty, K.; Corvera, S.; Czech, M.P. Unconventional myosin Myo1c promotes membrane fusion in a regulated exocytic pathway. *Mol. Cell. Biol.* **2004**, *24*, 5447–5458. [[CrossRef](#)]
34. Yip, M.F.; Ramm, G.; Larance, M.; Hoehn, K.L.; Wagner, M.C.; Guilhaus, M.; James, D.E. CaMKII-Mediated phosphorylation of the myosin motor Myo1c is required for insulin-stimulated GLUT4 translocation in adipocytes. *Cell Metab.* **2008**, *8*, 384–398. [[CrossRef](#)]
35. Chen, X.-W.; Leto, D.; Chiang, S.-H.; Wang, Q.; Saltiel, A.R. Activation of RalA is required for insulin-stimulated Glut4 trafficking to the plasma membrane via the exocyst and the motor protein Myo1c. *Dev. Cell* **2007**, *13*, 391–404. [[CrossRef](#)]
36. Huang, S.; Czech, M.P. The GLUT4 glucose transporter. *Cell Metab.* **2007**, *5*, 237–252. [[CrossRef](#)]
37. Tiwari, A.; Jung, J.-J.; Inamdar, S.M.; Nihalani, D.; Choudhury, A. The myosin motor Myo1c is required for VEGFR2 delivery to the cell surface and for angiogenic signaling. *Am. J. Physiol. Circ. Physiol.* **2013**, *304*, H687–H696. [[CrossRef](#)]
38. Kittelberger, N.; Breunig, M.; Martin, R.; Knölker, H.-J.; Miklavc, P. The role of myosin 1c and myosin 1b for surfactant exocytosis. *J. Cell Sci.* **2016**, *129*, 1685–1696. [[CrossRef](#)]
39. Sarshad, A.; Sadeghifar, F.; Louvet, E.; Mori, R.; Böhm, S.; Al-Muzzaini, B.; Vintermist, A.; Fomproix, N.; Östlund, A.-K.; Percipalle, P. Nuclear myosin 1c facilitates the chromatin modifications required to activate rRNA gene transcription and cell cycle progression. *PLoS Genet.* **2013**, *9*, e1003397. [[CrossRef](#)]
40. Hegan, P.S.; Ostertag, E.; Geurts, A.M.; Mooseker, M.S. Myosin Id is required for planar cell polarity in ciliated tracheal and ependymal epithelial cells: Myo1d is a determinant of planar cell polarity. *Cytoskeleton* **2015**, *72*, 503–516. [[CrossRef](#)]
41. Lechler, T.; Shevchenko, A.; Shevchenko, A.; Li, R. Direct involvement of yeast type I myosins in Cdc42-dependent actin polymerization. *J. Cell Biol.* **2000**, *148*, 363–374. [[CrossRef](#)] [[PubMed](#)]
42. Cheng, J.; Grassart, A.; Drubin, D.G. Myosin 1E coordinates actin assembly and cargo trafficking during clathrin-mediated endocytosis. *Mol. Biol. Cell* **2012**, *23*, 2891–2904. [[CrossRef](#)]
43. Maravillas-Montero, J.L.; López-Ortega, O.; Patiño-López, G.; Santos-Argumedo, L. Myosin 1g regulates cytoskeleton plasticity, cell migration, exocytosis, and endocytosis in B lymphocytes: Molecular immunology. *Eur. J. Immunol.* **2013**, *44*, 877–886. [[CrossRef](#)] [[PubMed](#)]
44. Arif, E.; Wagner, M.C.; Johnstone, D.B.; Wong, H.N.; George, B.; Pruthi, P.A.; Lazzara, M.J.; Nihalani, D. Motor protein Myo1c is a podocyte protein that facilitates the transport of slit diaphragm protein Neph1 to the podocyte membrane. *Mol. Cell. Biol.* **2011**, *31*, 2134–2150. [[CrossRef](#)]
45. Krendel, M.; Kim, S.V.; Willinger, T.; Wang, T.; Kashgarian, M.; Flavell, R.A.; Mooseker, M.S. Disruption of myosin 1e promotes podocyte injury. *J. Am. Soc. Nephrol.* **2008**, *20*, 86–94. [[CrossRef](#)]
46. Mele, C.; Iatropoulos, P.; Donadelli, R.; Calabria, A.; Maranta, R.; Cassis, P.; Buelli, S.; Tomasoni, S.; Piras, R.; Krendel, M.; et al. MYO1E Mutations and Childhood Familial Focal Segmental Glomerulosclerosis. *N. Engl. J. Med.* **2011**, *365*, 295–306. [[CrossRef](#)]
47. Wagner, M.C.; Blazer-Yost, B.L.; Boyd-White, J.; Srirangam, A.; Pennington, J.; Bennett, S. Expression of the unconventional myosin Myo1c alters sodium transport in M1 collecting duct cells. *Am. J. Physiol. Cell Physiol.* **2005**, *289*, C120–C129. [[CrossRef](#)]
48. Bi, J.; Chase, S.E.; Pellenz, C.D.; Kurihara, H.; Fanning, A.S.; Krendel, M. Myosin 1e is a component of the glomerular slit diaphragm complex that regulates actin reorganization during cell-cell contact formation in podocytes. *Am. J. Physiol. Ren. Physiol.* **2013**, *305*, F532–F544. [[CrossRef](#)]
49. Batters, C.; Arthur, C.P.; Lin, A.; Porter, J.; A Geeves, M.; A Milligan, R.; E Molloy, J.; Coluccio, L.M. Myo1c is designed for the adaptation response in the inner ear. *EMBO J.* **2004**, *23*, 1433–1440. [[CrossRef](#)]
50. Holt, J.R.; Gillespie, S.K.; Provance, D.; Shah, K.; Shokat, K.M.; Corey, D.P.; Mercer, J.; Gillespie, P.G. A chemical-genetic strategy implicates myosin-1c in adaptation by hair cells. *Cell* **2002**, *108*, 371–381. [[CrossRef](#)]
51. Donaudy, F.; Ferrara, A.; Esposito, L.; Hertzano, R.; Ben-David, O.; Bell, R.E.; Melchionda, S.; Zelante, L.; Avraham, K.B.; Gasparini, P. Multiple mutations of MYO1A, a cochlear-expressed gene, in sensorineural hearing loss. *Am. J. Hum. Genet.* **2003**, *72*, 1571–1577. [[CrossRef](#)] [[PubMed](#)]
52. Chen, Y.-D.; Yan, B.; Chalovich, J.M.; Brenner, B. Theoretical kinetic studies of models for binding myosin subfragment-1 to regulated actin: Hill model versus Geeves model. *Biophys. J.* **2001**, *80*, 2338–2349. [[CrossRef](#)]
53. Baek, J.-I.; Oh, S.-K.; Kim, D.-B.; Choi, S.-Y.; Kim, U.-K.; Lee, K.-Y.; Lee, S.-H. Targeted massive parallel sequencing: The effective detection of novel causative mutations associated with hearing loss in small families. *Orphanet J. Rare Dis.* **2012**, *7*, 60. [[CrossRef](#)] [[PubMed](#)]
54. Inouye, M.; Ripatti, S.; Kettunen, J.; Lyytikäinen, L.-P.; Oksala, N.; Laurila, P.-P.; Kangas, A.J.; Soininen, P.; Savolainen, M.J.; Viikari, J.; et al. Novel loci for metabolic networks and multi-tissue expression studies reveal genes for atherosclerosis. *PLoS Genet.* **2012**, *8*, e1002907. [[CrossRef](#)]
55. Tassopoulou-Fishell, M.; Deeley, K.; Harvey, E.M.; Sciote, J.; Vieira, A.R. Genetic variation in Myosin 1H contributes to mandibular prognathism. *Am. J. Orthod. Dentofac. Orthop.* **2012**, *141*, 51–59. [[CrossRef](#)]

56. Toyoda, T.; An, D.; Witzcak, C.A.; Koh, H.-J.; Hirshman, M.F.; Fujii, N.; Goodyear, L.J. Myo1c regulates glucose uptake in mouse skeletal muscle. *J. Biol. Chem.* **2011**, *286*, 4133–4140. [[CrossRef](#)]
57. Sun, R.; Wang, Y.; Jin, M.; Chen, L.; Cao, Y.; Chen, F. Identification and functional studies of MYO1H for mandibular prognathism. *J. Dent. Res.* **2018**, *97*, 1501–1509. [[CrossRef](#)]
58. Tyska, M.J.; Mackey, A.T.; Huang, J.-D.; Copeland, N.G.; Jenkins, N.A.; Mooseker, M.S. Myosin-1a is critical for normal brush border structure and composition. *Mol. Biol. Cell* **2005**, *16*, 2443–2457. [[CrossRef](#)]
59. Arif, E.; Solanki, A.K.; Srivastava, P.; Rahman, B.; Tash, B.R.; Holzman, L.B.; Janech, M.G.; Martin, R.; Knölker, H.-J.; Fitzgibbon, W.R.; et al. The motor protein Myo1c regulates transforming growth factor- β -signaling and fibrosis in podocytes. *Kidney Int.* **2019**, *96*, 139–158. [[CrossRef](#)]
60. Chase, S.E.; Encina, C.V.; Stolzenburg, L.R.; Tatum, A.H.; Holzman, L.B.; Krendel, M. Podocyte-specific knockout of myosin 1e disrupts glomerular filtration. *Am. J. Physiol. Ren. Physiol.* **2012**, *303*, F1099–F1106. [[CrossRef](#)]
61. Kim, S.V.; Mehal, W.Z.; Dong, X.; Heinrich, V.; Pypaert, M.; Mellman, I.; Dembo, M.; Mooseker, M.S.; Wu, D.; Flavell, R.A. Modulation of cell adhesion and motility in the immune system by Myo1f. *Science* **2006**, *314*, 136–139. [[CrossRef](#)]
62. Spielmann, M.; Hernandez-Miranda, L.R.; Ceccherini, I.; Weese-Mayer, D.E.; Kragesteen, B.K.; Harabula, I.; Krawitz, P.; Birchmeier, C.; Leonard, N.; Mundlos, S. Mutations in MYO1H cause a recessive form of central hypoventilation with autonomic dysfunction. *J. Med. Genet.* **2017**, *54*, 754–761. [[CrossRef](#)]
63. Ruppert, C.; Kroschewski, R.; Bähler, M. Identification, characterization and cloning of myr 1, a mammalian myosin-I. *J. Cell Biol.* **1993**, *120*, 1393–1403. [[CrossRef](#)]
64. Bähler, M.; Kroschewski, R.; E Stöfler, H.; Behrmann, T. Rat myr 4 defines a novel subclass of myosin I: Identification, distribution, localization, and mapping of calmodulin-binding sites with differential calcium sensitivity. *J. Cell Biol.* **1994**, *126*, 375–389. [[CrossRef](#)]
65. Köhler, D.; Struchholz, S.; Bähler, M. The two IQ-motifs and Ca²⁺/calmodulin regulate the rat myosin 1d ATPase activity: Regulation of Myo1d ATPase activity. *FEBS J.* **2005**, *272*, 2189–2197. [[CrossRef](#)]
66. Patino-Lopez, G.; Aravind, L.; Dong, X.; Kruhlak, M.J.; Ostap, E.M.; Shaw, S. Myosin 1G is an abundant class I Myosin in lymphocytes whose localization at the plasma membrane depends on its ancient divergent Pleckstrin Homology (PH) domain (Myo1PH). *J. Biol. Chem.* **2010**, *285*, 8675–8686. [[CrossRef](#)]
67. Gupta, P.; Gauthier, N.C.; Cheng-Han, Y.; Zuanning, Y.; Pontes, B.; Ohmstede, M.; Martin, R.; Knölker, H.-J.; Döbereiner, H.-G.; Krendel, M.; et al. Myosin 1E localizes to actin polymerization sites in lamellipodia, affecting actin dynamics and adhesion formation. *Biol. Open* **2013**, *2*, 1288–1299. [[CrossRef](#)]
68. Münnich, S.; Taft, M.H.; Manstein, D.J. Crystal structure of human myosin 1c—The motor in GLUT4 exocytosis: Implications for Ca²⁺ regulation and 14-3-3 binding. *J. Mol. Biol.* **2014**, *426*, 2070–2081. [[CrossRef](#)]
69. Dart, A.E.; Tollis, S.; Bright, M.D.; Frankel, G.; Endres, R.G. The motor protein myosin 1G functions in Fc γ R-mediated phagocytosis. *J. Cell Sci.* **2012**, *125*, 6020–6029. [[CrossRef](#)]
70. Swanson, J.; Johnson, M.; Beningo, K.; Post, P.; Mooseker, M.; Araki, N. A contractile activity that closes phagosomes in macrophages. *J. Cell Sci.* **1999**, *112*, 307–316. [[CrossRef](#)]
71. Nambiar, R.; McConnell, R.E.; Tyska, M.J. Control of cell membrane tension by myosin-I. *Proc. Natl. Acad. Sci. USA* **2009**, *106*, 11972–11977. [[CrossRef](#)] [[PubMed](#)]
72. Gérard, A.; Patino-Lopez, G.; Beemiller, P.; Nambiar, R.; Ben-Aissa, K.; Liu, Y.; Totah, F.J.; Tyska, M.J.; Shaw, S.; Krummel, M.F. Detection of rare antigen-presenting cells through T cell-intrinsic meandering motility, mediated by Myo1g. *Cell* **2014**, *158*, 492–505. [[CrossRef](#)] [[PubMed](#)]
73. López-Ortega, O.; Ovalle-García, E.; Ortega-Blake, I.; Antillón, A.; Chávez-Munguía, B.; Patiño-López, G.; Fragoso-Soriano, R.; Santos-Argumedo, L. Myo1g is an active player in maintaining cell stiffness in B-lymphocytes: Myo1g mediates cell stiffness in B-lymphocytes. *Cytoskeleton* **2016**, *73*, 258–268. [[CrossRef](#)] [[PubMed](#)]
74. Diz-Munoz, A.; Krieg, M.; Bergert, M.; Ibarlucea-Benitez, I.; Muller, D.J.; Paluch, E.; Heisenberg, C.-P. Control of directed cell migration in vivo by membrane-to-cortex attachment. *PLoS Biol.* **2010**, *8*, e1000544. [[CrossRef](#)]
75. Cordonnier, M.-N.; Dauzonne, D.; Louvard, D.; Coudrier, E. Actin filaments and myosin I alpha cooperate with microtubules for the movement of lysosomes. *Mol. Biol. Cell* **2001**, *12*, 4013–4029. [[CrossRef](#)]
76. Raposo, G.; Cordonnier, M.-N.; Tenza, D.; Menichi, B.; Dürrbach, A.; Louvard, D.; Coudrier, E. Association of myosin I alpha with endosomes and lysosomes in mammalian cells. *Mol. Biol. Cell* **1999**, *10*, 1477–1494. [[CrossRef](#)]
77. Salas-Cortes, L.; Ye, F.; Tenza, D.; Wilhelm, C.; Theos, A.; Louvard, D.; Raposo, G.; Coudrier, E. Myosin Ib modulates the morphology and the protein transport within multi-vesicular sorting endosomes. *J. Cell Sci.* **2005**, *118*, 4823–4832. [[CrossRef](#)]
78. McIntosh, B.; Pyrpassopoulos, S.; Holzbaur, E.L.; Ostap, E.M. Opposing kinesin and myosin-I motors drive membrane deformation and tubulation along engineered cytoskeletal networks. *Curr. Biol.* **2018**, *28*, 236–248.e5. [[CrossRef](#)]
79. Chugh, P.; Paluch, E.K. The actin cortex at a glance. *J. Cell Sci.* **2018**, *131*, jcs186254. [[CrossRef](#)]
80. Wang, F.-S.; Liu, C.-W.; Diefenbach, T.J.; Jay, D.G. Modeling the role of myosin 1c in neuronal growth cone turning. *Biophys. J.* **2003**, *85*, 3319–3328. [[CrossRef](#)]
81. Maravillas-Montero, J.L.; Gillespie, P.G.; Patiño-López, G.; Shaw, S.; Santos-Argumedo, L. Myosin 1c participates in B cell cytoskeleton rearrangements, is recruited to the immunologic synapse, and contributes to antigen presentation. *J. Immunol.* **2011**, *187*, 3053–3063. [[CrossRef](#)]

82. Schauer, K.; Duong, T.; Bleakley, K.; Bardin, S.; Bornens, M.; Goud, B. Probabilistic density maps to study global endomembrane organization. *Nat. Methods* **2010**, *7*, 560–566. [[CrossRef](#)]
83. Grossier, J.-P.; Xouri, G.; Goud, B.; Schauer, K. Cell adhesion defines the topology of endocytosis and signaling. *EMBO J.* **2014**, *33*, 35–45. [[CrossRef](#)]
84. McIntosh, B.B.; Holzbaaur, E.L.; Ostap, E.M. Control of the initiation and termination of kinesin-1-driven transport by myosin-Ic and nonmuscle tropomyosin. *Curr. Biol.* **2015**, *25*, 523–529. [[CrossRef](#)]
85. Tang, N.; Ostap, E. Motor domain-dependent localization of myo1b (myr-1). *Curr. Biol.* **2001**, *11*, 1131–1135. [[CrossRef](#)]
86. Collins, K.; Sellers, J.R.; Matsudaira, P. Calmodulin dissociation regulates brush border myosin I (110-kD-calmodulin) mechanochemical activity in vitro. *J. Cell Biol.* **1990**, *110*, 1137–1147. [[CrossRef](#)]
87. Kee, A.J.; Yang, L.; Lucas, C.A.; Greenberg, M.J.; Martel, N.; Leong, G.M.; Hughes, W.E.; Cooney, G.J.; James, D.E.; Ostap, E.M.; et al. An actin filament population defined by the tropomyosin Tpm3.1 regulates glucose uptake. *Traffic* **2015**, *16*, 691–711. [[CrossRef](#)]
88. Joensuu, M.; Belevich, I.; Rämö, O.; Nevzorov, I.; Vihinen, H.; Puhka, M.; Witkos, T.M.; Lowe, M.; Vartiainen, M.K.; Jokitalo, E. ER sheet persistence is coupled to myosin 1c-regulated dynamic actin filament arrays. *Mol. Biol. Cell* **2014**, *25*, 1111–1126. [[CrossRef](#)]
89. Soldati, T. Unconventional myosins, actin dynamics and endocytosis: A ménage à trois?: Myosins, actin dynamics and endocytosis. *Traffic* **2003**, *4*, 358–366. [[CrossRef](#)]
90. Jung, G.; Rimmert, K.; Wu, X.; Volosky, J.M.; Hammer, J.A. The dictyostelium carmil protein links capping protein and the Arp2/3 complex to type I myosins through their Sh3 domains. *J. Cell Biol.* **2001**, *153*, 1479–1498. [[CrossRef](#)]
91. Geli, M.I.; Lombardi, R.; Schmelzl, B.; Riezman, H. An intact SH3 domain is required for myosin I-induced actin polymerization. *EMBO J.* **2000**, *19*, 4281–4291. [[CrossRef](#)] [[PubMed](#)]
92. Manenschijn, H.E.; Picco, A.; Mund, M.; Rivier-Cordey, A.-S.; Ries, J.; Kaksonen, M. Type-I myosins promote actin polymerization to drive membrane bending in endocytosis. *eLife* **2019**, *8*, e44215. [[CrossRef](#)] [[PubMed](#)]
93. Wollenberg, R.D.; Taft, M.H.; Giese, S.; Thiel, C.; Balázs, Z.; Giese, H.; Manstein, D.J.; Sondergaard, T.E. Phenamacril is a reversible and noncompetitive inhibitor of Fusarium class I myosin. *J. Biol. Chem.* **2019**, *294*, 1328–1337. [[CrossRef](#)] [[PubMed](#)]